Journal of the Royal Society of Arts

NO. 4971 FRIDAY, 17TH FEBRUARY 1956 VOL. CIV

FORTHCOMING MEETINGS

WEDNESDAY, 22ND FEBRUARY, at 2.30 p.m. '*The British Glasshouse Industry*', by W. F. Bewley, C.B.E., D.Sc., V.M.H., Director, Glasshouse Crops Research Institute. Sir Edward Salisbury, C.B.E., D.Sc., F.R.S., V.M.H., Director, Royal Botanic Gardens, Kew, will preside. (The paper will be illustrated with lantern slides.)

WEDNESDAY, 29TH FEBRUARY, at 2.30 p.m. '*A New Concept in Theatre Design*', by Norman R. Branson, A.R.I.B.A. Tyrone Guthrie will preside. (The paper will be illustrated with a film, lantern slides and a working model.)

WEDNESDAY, 29TH FEBRUARY, at 6.30 p.m. DISCUSSION on '*Sport, with some Reference to the Growth of Professionalism*'. Opening Speaker: Denzil Batchelor. (Refreshments will be served from 5.45 p.m. See notice below.)

WEDNESDAY, 7TH MARCH, at 2.30 p.m. POPE MEMORIAL LECTURE. '*The Debt of Chemistry to Medicine*', by Sir Charles Dodds, M.V.O., M.D., D.Sc., F.R.C.P., F.R.S., Courtauld Professor of Biochemistry, University of London at Middlesex Hospital Medical School. Sir Cyril Hinshelwood, M.A., D.Sc., P.R.S., Dr. Lee's Professor of Chemistry, University of Oxford, will preside. (The lecture will be illustrated with lantern slides.)

THURSDAY, 8TH MARCH, at 5.15 p.m. HENRY MORLEY LECTURE. '*Recent Developments in Trade and Industry in Pakistan*', by His Excellency Mr. Mohammed Ikramullah, High Commissioner for Pakistan. The Right Honble. Lord Hailey, P.C., G.C.S.I., G.C.M.G., G.C.I.E., will preside. (Tea will be served from 4.30 p.m.)

N.B.—As it is expected that a large number of Fellows and their guests will wish to attend this meeting, admission will be by special ticket, application for which should be made to the Secretary of the Commonwealth Section not later than Monday, 27th February.

WEDNESDAY, 14TH MARCH, at 2.30 p.m. '*The Tourist Industry*', by J. G. Bridges, O.B.E., Director-General, The British Travel and Holidays Association. Sir John Elliot, M.Inst.T., Chairman, London Transport Executive, will preside. (The paper will be illustrated with a colour travel film of Britain.)

Fellows are entitled to attend any of the above meetings without tickets (except where otherwise stated), and may also bring two guests. When they cannot accompany their guests, Fellows may give them special passes, books of which can be obtained on application to the Secretary.

EVENING DISCUSSION MEETING

The second Evening Discussion Meeting will be held, on the same lines as the first meeting which took place on 1st February, on Wednesday, 29th February, at 6.30 p.m. The subject will be 'Sport, with some Reference to the Growth of Professionalism', and the discussion will be opened by Mr. Denzil Batchelor, Sports Editor of *Picture Post*, who has provided the following indication of the points on which he intends to elaborate:

> Sport has become one of the major public industries in Britain to-day, some £25 million a year being spent on betting; football attendances amount to approximately a million a week. In Europe victory or defeat in sport is a matter of political significance; Paris, after the war, considered Reg Harris the Englishman second only to Winston Churchill.
>
> The amount of attention devoted to sport in papers, broadcasting and television.
>
> The rise of professionalism. The difference between special payment for sporting performances as in America, and State sponsorship as in Russia. Is there any room for amateurs? The sports that remain truly amateur; is there any advantage in amateurism? Professionalism in the Olympic Games.

The Chair will be taken by Sir Harold Saunders, a Member of Council of the Society. A short report on the first Discussion Meeting will be published in the *Journal* in due course.

Tickets of admission are not required and Fellows are entitled to introduce two guests. Light refreshments will be served in the Library from 5.45 p.m.

THE ALBERT MEDAL

The Council are now considering the award of the Albert Medal of the Royal Society of Arts for 1956. They therefore invite Fellows of the Society to forward to the Secretary as soon as possible the names of such men of high distinction as they think worthy of this honour. The Medal was struck to reward

'distinguished merit in promoting the Arts, Manufactures and Commerce', and has been awarded as follows in previous years:

1864 Sir Rowland Hill
1865 His Imperial Majesty Napoleon III
1866 Michael Faraday
1867 Sir W. Fothergill Cooke and Sir Charles Wheatstone
1868 Sir Joseph Whitworth
1869 Baron Justus von Liebig
1870 Vicomte Ferdinand de Lesseps
1871 Sir Henry Cole
1872 Sir Henry Bessemer
1873 Michel Eugène Chevreul
1874 Sir C. W. Siemens
1875 Michel Chevalier
1876 Sir George B. Airy
1877 Jean Baptiste Dumas
1878 Sir Wm. G. (afterwards Lord) Armstrong
1879 Sir William Thomson (afterwards Lord Kelvin)
1880 James Prescott Joule
1881 Prof. August Wilhelm Hofmann
1882 Louis Pasteur
1883 Sir Joseph Dalton Hooker
1884 Captain James Buchanan Eads
1885 Sir Henry Doulton
1886 Samuel Cunliffe Lister (afterwards Lord Masham)
1887 HER MAJESTY QUEEN VICTORIA
1888 Professor Hermann Louis Helmholtz
1889 John Percy
1890 Sir William Henry Perkin
1891 Sir Frederick Abel, Bt.
1892 Thomas Alva Edison
1893 Sir John Bennet Lawes, Bt., and Sir Henry Gilbert
1894 Sir Joseph (afterwards Lord) Lister
1895 Sir Isaac Lowthian Bell, Bt.
1896 Professor David Edward Hughes
1897 George James Simons
1898 Professor Robert Wilhelm Bunsen
1899 Sir William Crookes
1900 Henry Wilde
1901 HIS MAJESTY KING EDWARD VII
1902 Professor Alexander Graham Bell
1903 Sir Charles Augustus Hartley
1904 Walter Crane
1905 Lord Rayleigh
1906 Sir Joseph Wilson Swan
1907 The Earl of Cromer
1908 Sir James Dewar
1909 Sir Andrew Noble
1910 Madame Curie
1911 The Hon. Sir Charles Algernon Parsons
1912 The Right Hon. Lord Strathcona and Mount Royal
1913 HIS MAJESTY KING GEORGE V
1914 Senatore (afterwards Marchese) Guglielmo Marconi
1915 Sir Joseph John Thomson
1916 Professor Elias Metchnikoff
1917 Orville Wright
1918 Sir Richard Tetley Glazebrook
1919 Sir Oliver Joseph Lodge
1920 Professor Albert Abraham Michelson
1921 Sir J. Ambrose Fleming
1922 Sir Dugald Clerk
1923 Major-General Sir David Bruce and Colonel Sir Ronald Ross
1924 H.R.H. THE PRINCE OF WALES
1925 Lieut.-Colonel Sir David Prain
1926 Professor Paul Sabatier
1927 Sir Aston Webb
1928 Sir Ernest (afterwards Lord) Rutherford
1929 Sir J. Alfred Ewing
1930 Professor Henry E. Armstrong
1931 H.R.H. THE DUKE OF CONNAUGHT AND STRATHEARN
1932 Frank (now Sir Frank) Brangwyn
1933 Sir William Llewellyn
1934 Sir Frederick Gowland Hopkins
1935 Sir Robert A. Hadfield, Bt.
1936 The Earl of Derby
1937 Lord (now Viscount) Nuffield

1938 HER MAJESTY QUEEN MARY
1939 Sir Thomas H. Holland
1940 John A. Milne
1941 President Franklin D. Roosevelt
1942 Field-Marshal J. C. Smuts
1943 Sir John Russell
1944 Sir Henry Tizard
1945 Winston (now Sir Winston) Churchill
1946 Sir Alexander Fleming and Sir Howard Florey
1947 Sir Robert Robinson
1948 Sir William Reid Dick
1949 Sir Giles Gilbert Scott
1950 Sir Edward Appleton
1951 HIS MAJESTY KING GEORGE VI
1952 Sir Frank Whittle
1953 Dr. E. D. (now Lord) Adrian
1954 Sir Ambrose Heal
1955 Dr. Vaughan Williams

EXHIBITION OF EUROPEAN MEDALS, 1930–1955

The Society's Exhibition of European Medals, 1930–1955, which, as has already been announced in the *Journal*, is at the moment touring the United Kingdom, is now on show at the Royal Scottish Museum, Edinburgh, where it was opened yesterday. The Right Honble. the Lord Provost of Edinburgh, Sir John G. Banks, C.B.E., J.P., presided at the opening ceremony which was performed by Sir William O. Hutchison, Hon. R.A., R.P., R.S.A., President of the Royal Scottish Academy. The Exhibition will remain in Edinburgh until 10th March, 1956.

Fellows will remember that the Exhibition, which was originally held at the Society's House last June, contains medals from 12 European countries, some coins, and a special exhibit of Coronation Medals provided by the Royal Mint.

THE SCIENCE OF BREWING

Three Cantor Lectures by

A. H. COOK, D.Sc., F.R.I.C., F.R.S.,

Assistant Director of the Brewing Industry Research Foundation

LECTURE I

Monday, 21st November, 1955

INTRODUCTION

In attempting to assess the scientific background of a particular industry, the picture which is eventually revealed depends to a large extent on whether that industry has been brought into existence only as a result of advancing knowledge or whether it employs primarily an accumulation of empirical experience. Brewing, with its background of at least six and perhaps as much as ten thousand years' practice, falls obviously into the latter category and it would therefore be misleading to suggest that much of that practice takes its present form because of science. Nevertheless brewing has close associations with some of the most important advances in science and it continues to provide the starting point for many fundamental studies. It may be recalled, for instance, that Leeuwenhoek's first observations with his newly elaborated compound microscope were with yeast and these in fact signalled the origin of microbiology. Again, Pasteur was probably the first to perceive some of the important medical implications of microbiological life as a result of his studies of wine and beer fermentations, while it was brewing studies which led Kjeldahl to devise the method of determining nitrogen associated with his name, and investigations in the same field which led Sørensen to the now universally adopted concept of pH. With this background in mind, therefore, it seemed appropriate to use the present opportunity to trace within the limitations of three lectures some of the contributions which the study of brewing and brewing materials have recently made to natural knowledge. This approach must not be taken to imply that science is destined to remain of little positive value to the brewer. Economic circumstances among others are enforcing changes in the barley field and the maltings, in the hop garden and hop oast, and in the brewery. Thus, in the case of barley a potent factor is the increased demand to support an expanded national animal stock. New varieties of hops are also receiving widespread attention, particularly with regard to such factors as mechanized hop-picking and the need to develop and maintain disease-resistant varieties of hops. The formulation of these changes and their prompt acceptance clearly depends largely on the measure of understanding as to how far the new materials meet the basic requirements of the old ones. In this sense it seems likely that the science of brewing has come to the cross-roads. Behind stretches tradition with nevertheless, in latter years, an ever-increasing exchange between technology and exact science.

In front, however, the vista reveals to an increasing extent a brewing industry increasingly attuned to a detailed scientific appraisal of all its constituent processes. Moreover, it seems likely that a keener awareness of the possibilities of microbiological industry may lead to the association of brewing with a number of daughter industries which may, in respect of size and importance, come in due course to dwarf the production of beer itself.

Undoubtedly the very traditions of brewing have in the past unwittingly served to restrict this broader vista. It is interesting to note that when Graham delivered in 1875 the first Cantor Lectures devoted to the present subject, the account which he was able to give of the scientific basis of the industry was so detailed that, considering the level of knowledge of that time, brewing must have been of all industries the one best informed of its technical background. However, it is undeniably true that for most of the succeeding half-century there was an uneasy fear in the industry that any pronounced leaning towards science might detract from the art of brewing and might lead to suspicion in the public mind. It is recorded for example, in another lecture to this Society, that when a certain brewery installed a laboratory the management insisted at a late stage that the word 'Laboratory' should be removed from over the main entrance because it could be seen from the public street. Happily this attitude is everywhere steadily giving way to a more enlightened outlook, as is evidenced by the establishment in this country of the Brewing Industry Research Foundation. That the latter is indeed the first central scientific establishment in the whole world financed entirely by the industry to carry out research for the advancement of brewing as a whole is surely tangible proof of an imaginative leadership which offers both opportunity and challenge to the diverse powers of modern science.

THE BREWING PROCESS

It may assist those who are unfamiliar with the industry to review the outlines of the whole brewing process. The latter may be regarded as starting with barley which, as a result of steeping in water, germination and finally kilning, is converted into malt. By mashing, that is by treating with hot water under controlled conditions, malt, either alone or supplemented by wheat, maize, rice or sugar, yields a sweet extract or wort. The latter is boiled with hops whereby a variety of changes in the composition of the wort are brought about. It is hopped wort which is fermented by the action of yeast to give a green beer which is allowed to mature. The conditioned beer is finally treated in various ways according to its type and according to whether it is to be bottled or sold in cask as finished beer. These stages, together with certain additional data, are summarized in Figure 1.

It will not be possible in view of limitations of time and space to consider in detail many of the technical variations of these processes. It must suffice at this point to mention that malting is commonly carried out in this country according to a system, the characteristic feature of which is the use of floors on which the barley is allowed to germinate; during this stage it is turned from time to time

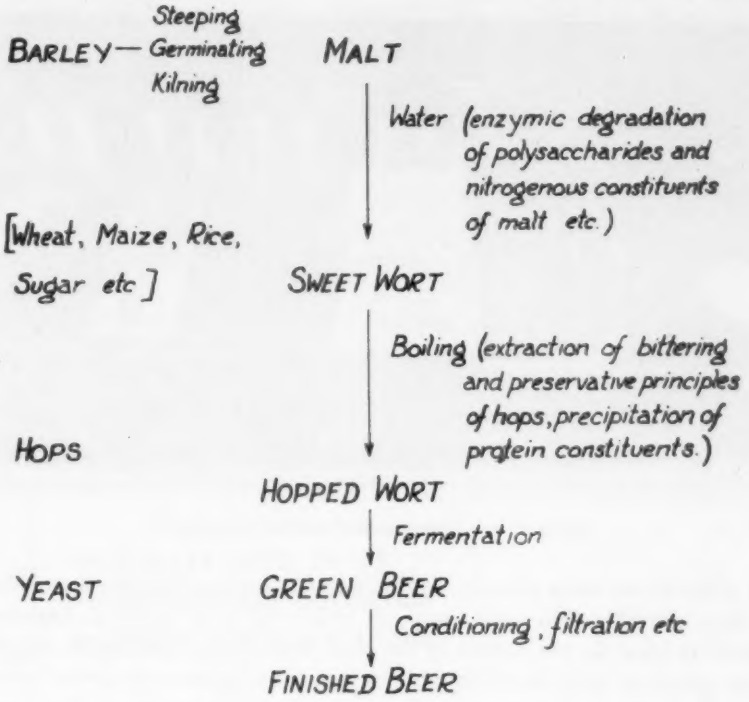


FIGURE 1. *The brewing process*

either mechanically or by hand to secure even growth. Alternative systems employ slowly rotating drums or boxes through which air can be passed to assist the respiration of the young barley plants. About 10 million cwt. of barley per year are malted for brewing in Great Britain but it is interesting, in view of certain developments mentioned later, to note that currently the total usage of barley for malt for all purposes here amounts to no less than 35 million cwt.

Figure 2 shows part of the unique, specially designed and self-contained experimental malting apparatus at the Brewing Industry Research Foundation, which is capable of satisfactorily malting ½ lb. of barley as compared with the thirty tons which may be taken as an average batch in a full-scale plant.

Coming to brewing itself, the variations may be said to resolve themselves primarily round top-fermentations, such as are characteristic of brewing in Great Britain, and bottom-fermentations which distinguish lager brewing as carried out extensively on the Continent and in America. The names refer to the behaviour of the yeast in rising to the surface or sinking in the fermenting liquid. These two systems are associated with somewhat different detailed procedures, for instance, for the production of wort and the maturing of beer. The precise temperatures, for example, at which mashing, fermentation and maturation are carried out differ substantially between top-fermentation and lager brewing. There are in addition numerous important variations within

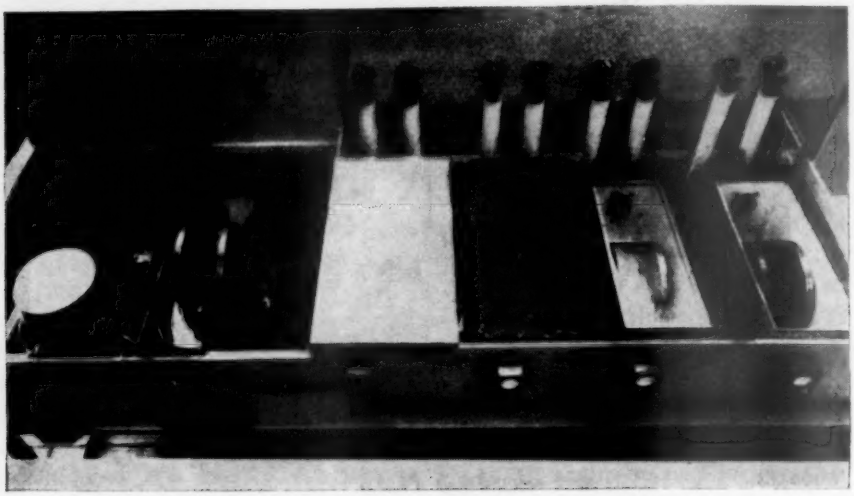


FIGURE 2. *Experimental malting apparatus*

each of these two main systems. In this country, for instance, the fermentations are often carried out in open vessels in which the yeast forms a characteristic undulating layer on the surface of the beer, whereas in other cases the Burton Union system is used, in which the 'top' yeast is continuously carried over into a subsidiary vessel. Varying procedures are employed, moreover, for removing the main bulk of yeast at the end of the fermentation. Nevertheless, many of the scientific principles involved are common to all systems of brewing and it is with such general considerations that the following discussion is mostly concerned.

BARLEY AND MALT

Barley is, quantitatively at least, the most important raw material of brewing and its composition calls therefore for detailed consideration. It must be borne in mind at the same time that the composition of barley often varies according to its variety, agricultural history and even with different storage conditions. However, even if barleys of unvarying composition were attainable, they would be subject to variable processes of malting and of mashing so that the original composition may not be so decisive a factor as it might first appear. Obviously, however, a knowledge of the composition is desirable if the variability of the subsequent processes is to be purposefully directed in the best manner. Interest naturally centres in this respect on newer varieties, for considerable effort is being devoted to the breeding of improved barleys both in this country, notably at the National Institute for Agricultural Botany, and abroad where the work of the Barley Improvement Association in the United States must be specially mentioned. The development of new hybrid barleys in this country at least is dictated primarily by a pronounced shift in the demand for high-yielding feed barleys as compared with those grown earlier more specifically for malting and brewing purposes. It will be noted from Figure 3 that while the absolute

consumption of brewing-malting barley in this country has shown no very decided trend over a considerable number of years the relative consumption has dropped from rather more than fifty per cent to slightly less than twenty per cent of the total home-produced and imported barley over a relatively short period. It is increasingly difficult, therefore, to ensure for purely malting barley even a premium price which is economic both to the farmer and to the maltster and brewer. The tendency is consequently to concentrate on dual purpose barleys, even though they may call for modified malting and brewing methods, and it is precisely at this point that science is playing an increasingly important part.

Hybrid varieties of barley should be first selected so that only those of acceptable agricultural characteristics are later produced in sufficient quantity for their malting and brewing quality to be assessed. There is clearly a need for these latter properties to be evaluated in an objective manner and to this end it is desirable to be able to define barley composition in fundamental terms.

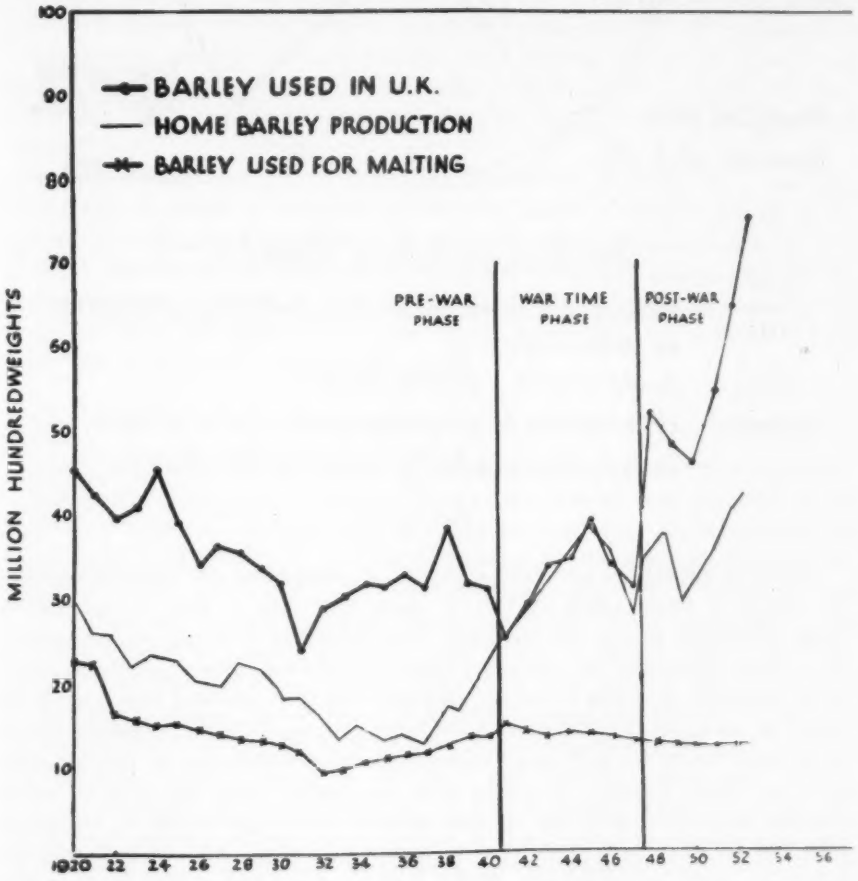


FIGURE 3. *Production and use of barley*

Actually it will be apparent that the attempt raises many difficult questions, so that this one problem alone brings one to the frontiers of science at many different points.

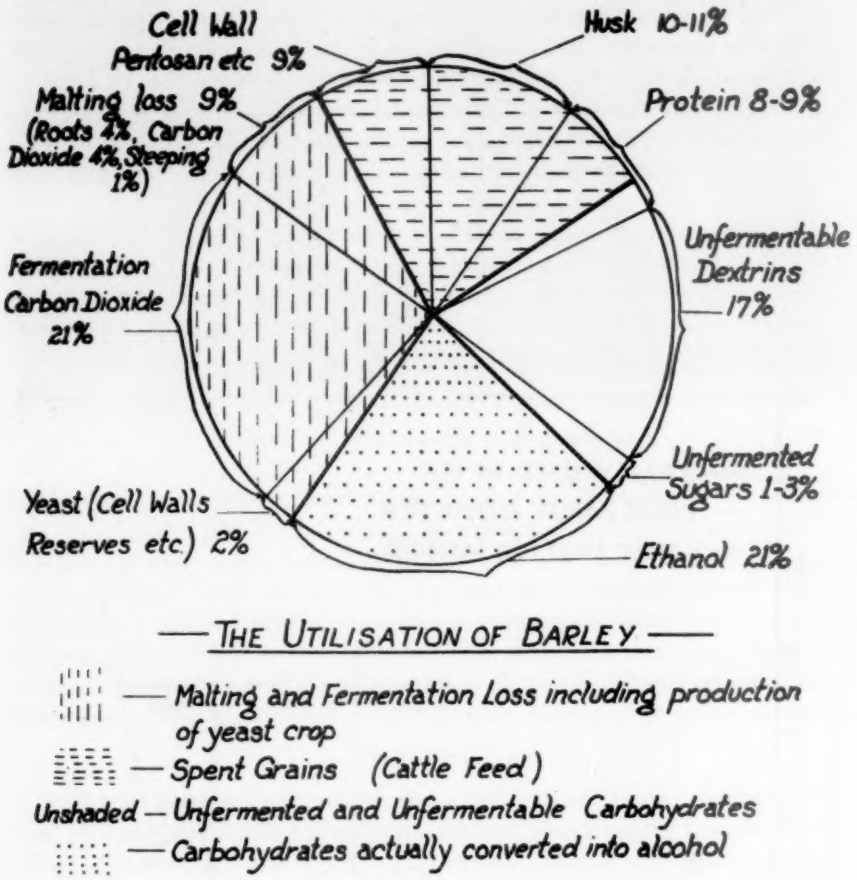

Cell Wall
Pentosan etc 9%
Husk 10-11%
Malting loss 9%
(Roots 4%, Carbon Dioxide 4%, Steeping 1%)
Protein 8-9%
Unfermentable Dextrins 17%
Fermentation Carbon Dioxide 21%
Unfermented Sugars 1-3%
Yeast (Cell Walls Reserves etc.) 2%
Ethanol 21%
THE UTILISATION OF BARLEY
— Malting and Fermentation Loss including production of yeast crop
— Spent Grains (Cattle Feed)
Unshaded — Unfermented and Unfermentable Carbohydrates
— Carbohydrates actually converted into alcohol

FIGURE 4

Sufficient superficial findings to provide a background are brought together in Figure 4 from which it will be seen that only a part of the barley grain ultimately passes into the beer. The brewer is of course not concerned solely with producing the maximum amount of alcohol from a given amount of grain, for many qualities of flavour and appearance in the final beer have to be taken into account. On the other hand there appears from the following *résumé* substantial scope for still more economical and efficient use of the available material. From Figure 4 it will be seen that eight to nine per cent of barley consists of protein and that of this quantity about two-thirds is ultimately removed together with husk (ten to eleven per cent) and cell-wall material (nine per cent), as cattle-feed in the form of spent grains after malting and

mashing. Approximately nine per cent of the barley is lost during malting by dissolution of carbohydrates during steeping, by respiration, and by the removal of malt rootlets. A further proportion of the original barley, now in the form of malt, is expended in the production by fermentation of carbon dioxide (21 per cent) and of new yeast cells (two per cent) which also make use of some of the protein solubilised during malting and mashing. Still about twenty per cent of the original barley substance remains in the final beer, either as unfermentable dextrin or as unfermented sugars. The net result is that, of the barley employed in the complex process of brewing, only about 21 per cent is actually converted into ethyl alcohol. Obviously these approximate figures are to some extent inextricably bound up with the chemical reactions inevitably involved. For instance the production of ethanol from glucose by the overall reaction represented by the equation

$$C_6H_{12}O_6 \longrightarrow 2CO_2 + C_2H_5OH$$

inevitably involves about as much carbon dioxide being lost as ethyl alcohol is formed. In other respects, however, notably in connection with the unfermented carbohydrates and the losses associated with malting, it would appear that there is possibly room for improving the efficiency of utilization of barley without detracting from the desirable characteristics either of the derived malt or of the finished beer.

Attention must obviously primarily be focused for the purposes of brewing on the carbohydrates, as these are the ultimate source of most if not all of the alcohol produced by fermentation. However, nitrogenous components are also of great importance, as they include enzymes which are associated with the various changes induced during malting and mashing, together with as yet ill-defined materials largely responsible for yeast nutrition, the stability of beer foam and commercially undesirable hazes.

GENERAL PROPERTIES OF BARLEY

It will be convenient to consider in the first place certain general characteristics of barley which have not in the past been readily related with precision to its detailed composition. Because of the latter circumstance, the assessment of a barley as to its suitability for malting is often based to some extent on subjective tests. For instance visual examination can, with experience, give in many instances reliable information on the variety, its freedom from mould contaminants, its moisture content and other characteristics. Often, of course, such inspection gives in a general way information which can now be assayed more accurately by specially designed laboratory tests. Thus the desirable fineness of skin is a general reflection of a lower husk content and therefore of a probably higher carbohydrate content; but it is impossible and possibly unnecessary in the present lectures to review in detail such routine laboratory estimations used to supplement visual examination of barleys. It must suffice to remark that they include determinations of moisture-content, nitrogen-content and the weight of a thousand corns and that by such means, employing empirically determined

relations, the amount of extract obtainable from a derived malt can usually be predicted with considerable accuracy. Figures for all the brewing materials refer in the present review to their computed dry weight, which in the case of barley usually entails allowing for a water-content of 12 to 14 per cent. The precise figure is difficult to define because although the water-content of a barley stored normally is an equilibrium figure depending on the humidity of the surrounding air, attempts to determine it absolutely lead to apparently irreversible protein changes and loss of some of the ability to germinate, as the water content is reduced below about ten per cent. Reverting then to routine laboratory estimations, perhaps the present opportunity will be better used by reviewing selected aspects of recent research in this connection and here mention must first be made of important considerations with regard to the germination of the barley which is indispensable for its conversion into malt.

GERMINATION OF BARLEY

It often happens that barley, especially soon after harvesting, fails to germinate fully under malting conditions. Obviously this may be due in part at least to an undesirable proportion of the corns being dead and it is customary therefore to determine the *germinative capacity* of the grain. Now if the corn is actually alive it can be brought into active germination by damaging the outer husk (Figure 5), for example, by piercing it with a pin, or by removing it altogether. Its dormancy therefore is associated with the outer layers, and while the phenomenon of dormancy may be partly associated with a physical impermeability of these layers to water, it is also undoubtedly related as well to chemical factors. Indeed, the steeping of barley results in the removal in the steeping liquor of a variety of substances, including vanillic acid, which can be shown to retard the germination

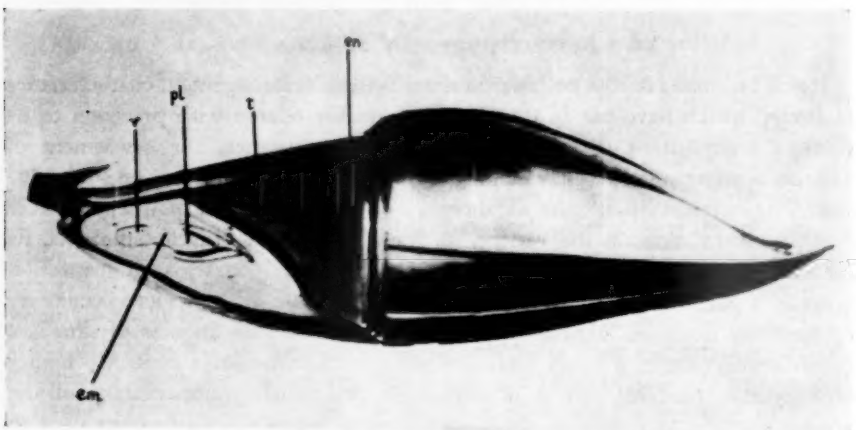


FIGURE 5. *Detailed structure of the barley corn*
r = radicle em = embryo pl = plumule
t = testa en = endosperm

of barley and other seeds[1]. Clearly the process of steeping barley is concerned with much more than securing the direct imbibition of water as was at one time supposed. Normally the young barley plant develops both a rootlet and a shoot. Experience has taught that under normal conditions it is satisfactory for the production of malt to arrest growth by kilning when the shoot or acrospire has grown about two-thirds of the length of the grain. This degree of growth is accompanied by a dense mat of rootlets which have subsequently to be rubbed off and removed altogether, so accounting for some of the usual malting loss. Now one effect of certain of the inhibitors of germination already mentioned is to suppress more particularly the growth of rootlets. The deliberate use of these materials can therefore lead to 'malt' associated with a reduced malting loss and when due account is taken of other factors such processes may well be of substantial economic advantage. The experimental overcoming of dormancy in barley by peeling the corns is naturally tedious, and it was a remarkable finding therefore that the husks can be conveniently removed by soaking the corns in strong or even concentrated sulphuric acid without harming the germ which is obviously adequately protected[2]. This convenient means of removing the protective husk from barley makes possible for the first time a number of investigations of profound interest both for brewing and for science in general. For instance, it is possible to produce malts without husk and to examine the properties of the ultimate beers. Investigations of this kind, such as are now in progress at the Brewing Industry Research Foundation, are of special significance because of suggestions that tannins derived from the husk are in part associated with the formation of undesired haze. Again, the germ or embryo of the barley merely lies alongside the starchy endosperm after stripping the husk from the grain. It is in fact easily detached and thus becomes available to facilitate the study of the life-process itself.

Reverting to the subject of germinative behaviour, a given sample of barley should of course have a high germinative capacity, that is to say, the proportion of corns ultimately capable of germinating should approach a hundred per cent. As mentioned above, the actual germination under laboratory conditions arbitrarily selected so as to provide as far as possible a useful comparison with large-scale malting conditions may fall far below that figure, so that its *germinative energy* is said to be low. Even if the corns should all germinate ultimately, any substantial irregularity from seed to seed in this respect is of serious concern to the maltster, although it may not be of great consequence to the farmer. This is because the whole 'piece' of the barley necessarily receives the same limited time on the floor, with the result that those corns which are slow to germinate have correspondingly less time for development with the result that the final malt is of uneven quality.

Recent studies of a wide range of barleys of low germinative energy[3] has brought to light two distinct aspects of their dormancy. The first of these is reflected in a deep-seated unwillingness to germinate even under wide variations of the normal conditions. This type of dormancy is commonly present in freshly harvested barleys, but is fortunately relatively short-lived and usually disappears

from the dried barley within a few weeks. It has been discovered that its disappearance can be hastened by adding to the water used for steeping a small quantity of any of a selected group of simple thiols, and here a very dilute (about 0·05 per cent) solution of hydrogen sulphide is probably the cheapest and most convenient. Under these conditions such a barley may then be malted even when fresh from the field and large-scale tests show that the product is thereby considerably improved both in respect of appearance and of the amount of extract which the brewer can obtain with it. Figure 6 reveals the improved behaviour of a particular sample of barley treated in this way.

Analysis	*Treatment*		
	None	0·025% H_2S *at maltings*	0·025% H_2S *in experimental malting apparatus*
Germination, %	79	89	95
Hot water extract (%, dry) ...	93·8	97·9	98·7
Cold water extract (%, dry) ...	14·9	15·2	16·3
D.P.	38·8	36·0	38·8
1000-corn weight (g, dry) ...	33·8	34·0	33·7
Malting loss (%, dry)	7·4	6·9	7·7

FIGURE 6. *The effect of hydrogen sulphide in germinating barley*

Barleys exhibiting the second aspect of dormancy are distinguished by the property of taking up water unduly rapidly on steeping and these are said to be water-sensitive. The deleterious effects appear to arise because while an uptake of about 35 per cent of water is required to initiate germination, the actual development of the growth process is sometimes relatively slow. In such cases the uptake of water nevertheless continues and may indeed reach a point where the growth is brought to an end. In other words, the germ is drowned by the rapidity of the uptake of water which is, on normal germination, soon brought under control by the activity of the young plant. From the maltster's viewpoint this behaviour is doubly troublesome, because while the quickly germinating corns will provide fully formed malt the slower germinating ones may well not develop at all. An appreciation of this factor now enables a barley to be steeped advantageously according to its particular response to water as judged by laboratory tests. The details of the steeping involve essentially soaking the barley, to permit only sufficient moisture to be taken up to provide for germination, and then draining off the water so that the initial stages of germination can proceed to completion before the remaining water required is added.

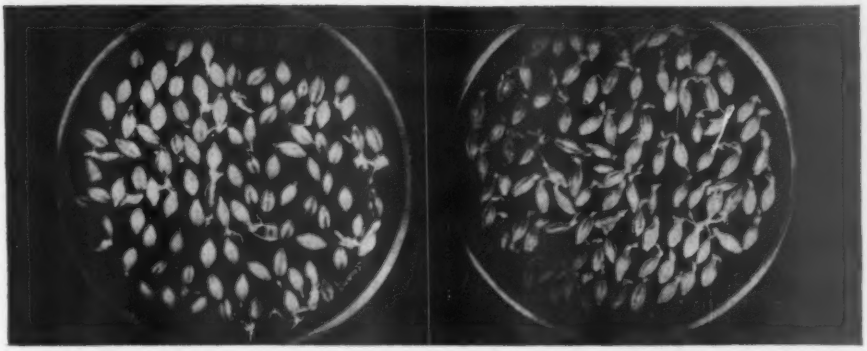


FIGURE 7. *Effect of steeping procedure on germination of barley*

A comparison between a typical barley germinated in this way and one which has received only straight-forward soaking is seen in Figure 7, which clearly shows the more regular germination brought about by controlling the access of water to the germinating seeds in the second photograph.

THE CARBOHYDRATES OF BARLEY AND MALT

Coming to the question of the chemical make-up of barley, the general composition of its carbohydrates has been the subject of considerable discussion[4]. Some of this is, however, necessarily inexact because apart from glucose, fructose, sucrose, maltose, isomaltose and raffinose it is often concerned with materials which lack the unvarying characteristics of chemical entities. Nowhere in the present connection is this more important than in the case of the major carbohydrate reserve of barley, namely starch. Unfortunately, despite much careful work[5] no one standardized preparation has yet been generally accepted as representing the whole starch of barley, but subject to this inevitable reservation it may be taken that about 62 per cent of the dry grain consists of material which can loosely be termed starch. Most of the significant work on the structure of starch in general has been carried out on potato or maize starch and very little has been done directly on barley or malt. The essential findings depend upon the separation from such whole starches of various origins of a component termed amylose, by treating a gelatinized suspension in hot water with one of a variety of precipitants such as butyl alcohol[6]; from the supernatant liquor a second material termed amylopectin is then recovered. Starch is thus heterogeneous and it is probable that the heterogeneity is generally increased by other factors. For instance, the starches of whole grain are generally associated with small but varying quantities of phosphorus, present as polysaccharide phoshate, of nitrogen, of minerals and of so-called hemicellulosic materials characterized by their content of pentoses. As already mentioned, practically no studies of this kind have been conducted on barley starch but the few structural investigations which do bear on the question suggest that barley starch is closely comparable

with starches of other origins[7] It is nevertheless advisable to bear in mind constantly that the following considerations probably refer only to the extreme types of structures present in barley starch and that the preparative procedures used were necessarily non-specific and doubtless tend to obscure the considerable overlap which must occur between the various structures depicted.

By taking account of the great capacity of amylose as compared with amylopectin to afford a deep blue adduct with iodine[8], it has been estimated that usually between eighteen and twenty per cent of whole starch consists of amylose. The amylose-content of ripe barley starch is about twenty per cent and is thus comparable with many other starches. Now immature corn contains starch with a much lower percentage (five to seven per cent) of amylose. If the same be true of barley the amylose content may be a valuable reflection of maturity, in view of the act that neither the whole starch nor the simpler saccharides appear to undergo any great change over an extended pre-ripening period[10].

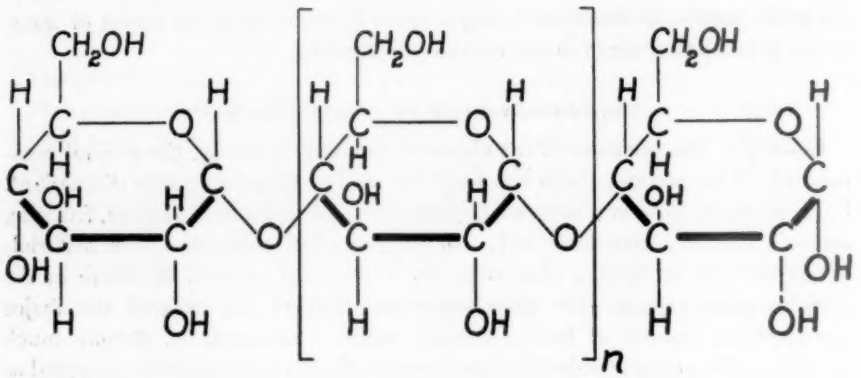


FIGURE 8. *Postulated structure of amylose*

The molecule of amylose appears to consist mainly of chains of several hundreds of glucose units each linked by α–1:4 bonds to give a total molecular weight of the order of 200,000. This general view is supported by end-group determinations either by methylation followed by estimation of the yield of tetramethyl-D-glucose formed by subsequent hydrolysis, or by estimating the yield of formic acid produced on oxidation with periodate.

Similar end-group determinations carried out on amylopectin, particularly by means of periodate[11] indicate one end-group for each 25 to 27 glucose units. Recalling therefore the approximate and average nature of such results, amylopectin obviously has a much more branched structure than amylose. For diagrammatic purposes, three extreme structures (*a*, *b* and *c*) have been envisaged though it should be emphasized that it is unnecessary to postulate any one type to the exclusion of the others. In each case the pattern is to be

regarded as extended to give a molecular weight of 1,000,000 or possibly more:

FIGURE 9. *Diagrammatic representation of amylopectin structure (G=Glucose unit)*

These two main starch components are quantitatively the most significant elements of barley and, after some modification, ultimately malt from the point of view of fermentable material. Perhaps the most comprehensive survey of the other carbohydrate constituents in approximate structural terms is provided by the work of G. Harris and I. C. MacWilliam[18] for a specified (Plumage-Archer) barley. The results together with others to which reference will be made later are summarized in Figure 10. Further (as yet unpublished) results with certain other varieties (Carlsberg and Proctor) reproduce the general picture conveyed by the results quoted. The analyses are based on long series of chromatograms of arbitrarily chosen groups of the carbohydrates concerned. A typical chromatogram on paper is reproduced in Figure 11. This method of chromatography on paper is of wide application in the quantitative examination of the carbohydrates not only of barleys and malts but also of worts and beers. Further examples of its use are included below and it seems likely that the method will find increasing routine use in malting and brewing laboratories as it offers a sensitivity and refinement not attainable by any other general approach. The extent to which the simpler carbohydrates (shown

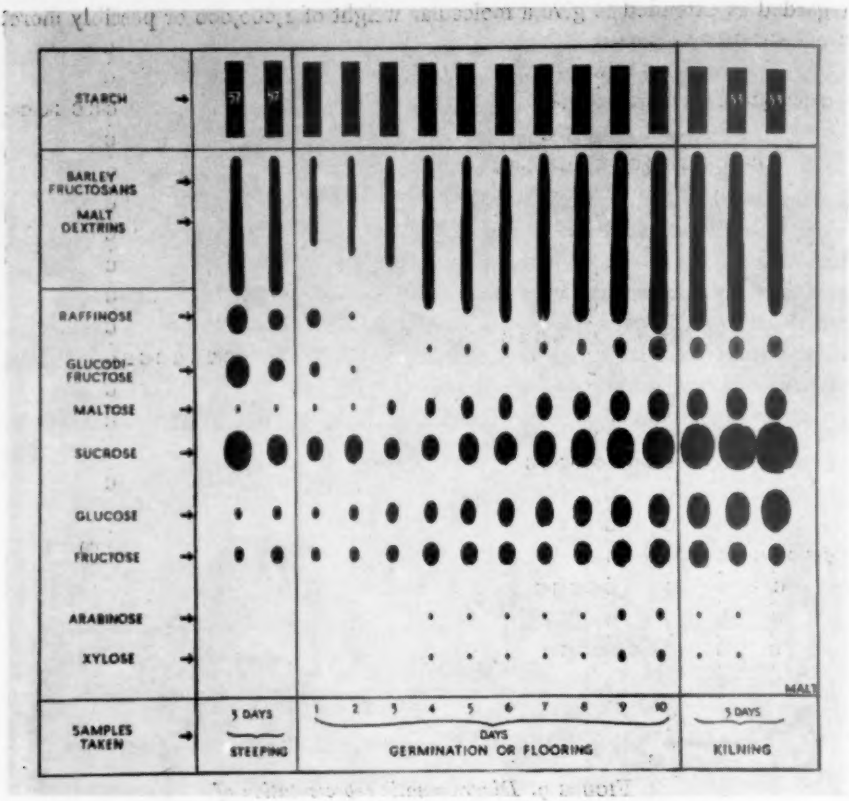


FIGURE 10. *Carbohydrate composition of barley and malt*

in Figure 10) represent steps in the synthesis or breakdown of starch or other complex polysaccharides, or the extent to which even in resting barley the picture is a static one, remains unknown. The question is of practical import, for instance, in disposing of the problem of dormancy and of great general interest in connection with the mechanism of germination. For example, an early result of active germination is the utilization of the simple sugars by respiration so that the grain is temporarily depleted of them. Later, as the growing plant achieves a wider organization of its activities, amylolytic enzymes are produced by the embryo and thence by a mechanism which remains obscure are more widely distributed throughout the grain so that for a time the content of simpler carbohydrates is dynamically restored from the reservoir of starch.

So far, attention has been directed chiefly to the carbohydrates of barley, but of more direct interest from the brewer's standpoint is perhaps the comparable composition of malt. Here a salient result of malting is a decrease of about five per cent in the starch content, although other changes such as the relatively large though absolutely small increase in sucrose, particularly during kilning of the malt, may in time prove to be of significance. It is noteworthy that,

although a great deal of maltose appears during mashing, little is produced up to this point. In other words the young plant derives its energy almost entirely by the indirect consumption of starch. However, the seemingly small decrease in starch probably masks a surprisingly extensive change for Aspinall, Hirst and McArthur[13], have shown that the amylopectin of malt has one non-reducing end-group to an average of only 18 glucose units compared with a ratio of about 1:26 in barley. Taking into account the relative proportions (about 1:4) of amylose and amylopectin in barley, the overall loss of starch during malting by respiration and root growth is less than would result by the exclusive metabolism of the ends of the amylopectin chains to such an extent that only the original chains, decreased in length by one-third, remained. Pending more accurate estimates of the ratio of amylose and amylopectin in malt it must be concluded therefore that at least a proportion of new starch is actually synthesized during malting. Some fragments of the chain-ends must either be linked to form new amylose molecules or are assembled to form new amylopectin molecules. This conclusion does not, of course, exclude the possibility that the larger residues of the original starch components are simultaneously rearranged in new patterns, but it would seem that much additional research, possibly with the aid of isotopically labelled starch, will be required before these and other fascinating questions can be more fully explored.

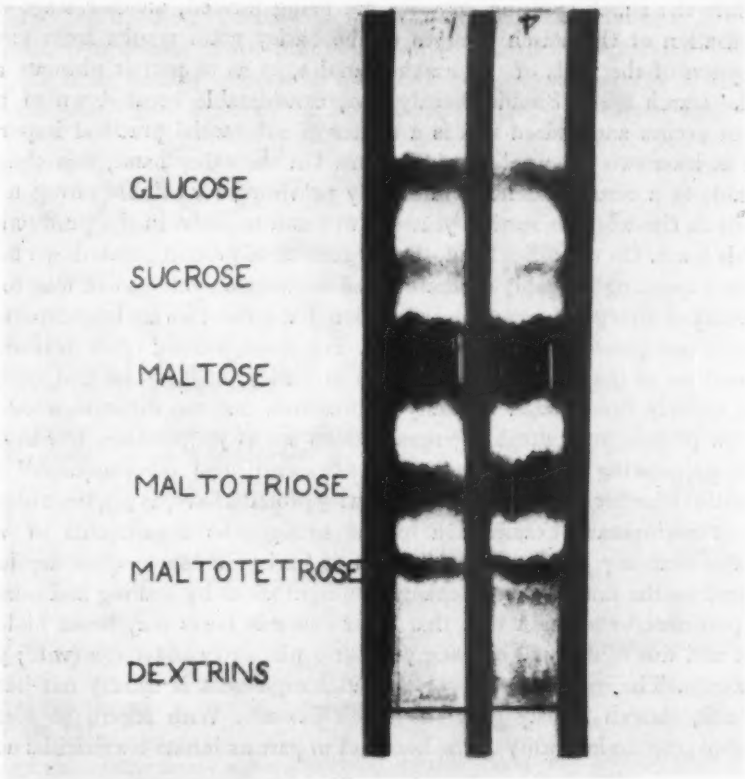


FIGURE 11. *A typical chromatogram on paper*

LECTURE II

Monday, 28th November, 1955

MASHING

The foregoing account provides a provisional picture of barley and malt starch as perhaps having identical amylose portions but differing in that the amylopectin of malt is more 'stubby' than that of barley.

The main object of malting is essentially to ensure that the carbohydrates shall be readily converted into the desired fermentable carbohydrates in the wort. This is ultimately achieved through the agency of enzymes. Those which are concerned occur to some extent in the original barley, while at least one important enzyme is formed by the young barley plant during malting itself. There is no doubt that the complete enzymic picture will prove to be extremely complex but certain useful outlines may be discerned as follows. Barley itself affords a measure of starch-splitting enzyme (β-amylase) but during malting a second major enzyme (α-amylase) is also formed. The two may be distinguished by the different rates at which they are inactivated by heat and by taking account of this and other characteristics they have been separated, possibly incompletely, and their properties studied.

While the starch-splitting enzymes are being formed, another stage of the mobilization of the starch reserves of the barley grain results from enzymic disruption of the walls of the starch granules, so as to permit ultimate attack on the starch itself. Simultaneously also, considerable break-down of barley protein occurs and indeed this is a matter of substantial practical importance from at least two balanced considerations. On the other hand, this change is desirable to a certain extent in ultimately providing for soluble nitrogen compounds in the wort, to further yeast growth and to assist in the production of a stable foam. On the other hand, the occurrence of protein break-down beyond a point depending probably on the over-all brewing conditions can lead to such a quantity of nitrogen compounds in solution that difficulties are later experienced by the development of undesired hazes. For these, among other reasons, the composition of the nitrogenous fractions of barley, malt, worts and beers are being actively investigated in many laboratories, but the difficulties are such that few positive reproducible generalizations are as yet possible. For instance, recent painstaking electrophoretic and ultracentrifugal investigations[14] make it doubtful whether any significant pure barley proteins have, as yet, been obtained while a preliminary examination of the nitrogenous constituents of wort[15] indicates that any characteristic features of barley in this respect are largely obscured by the multiplicity of changes brought about by malting and mashing. It is pertinent to mention here that other hazes in beers may be of biological origin and due to residual brewing yeast or to the entry of foreign (wild) yeasts or bacteria. The mere presence of the latter organisms is usually not harmful to health, though it may give rise to off-flavours. With regard to bacterial infections, the slight acidity of the beer and in part its innate bactericidal action,

due to the presence of traces of hop products, combine to lessen the entry of bacteria.

Malting is thus accompanied by the production of a variety of enzymes until the increasing momentum of their development is brought almost entirely to a standstill by kilning the green malt. The cessation of enzymic activity is partly a result of inactivation by heat and partly a consequence of the removal of water. By taking advantage of these facts a range of malts can be prepared having different amylolytic activities and indeed the content of a malt with respect to its starch-splitting enzyme is often an important index to its usefulness. For instance, a malt cured at a low temperature and retaining a high proportion of its original enzymic activity is particularly suited to the preparation of malt extract; one cured at an intermediate temperature is usually favoured for the production of pale ale in view of the delicate flavour so imparted to it, while a dark roasted malt largely or even entirely devoid of enzyme activity is particularly valued in the brewing of stout.

The mashing process consists essentially in adding the crushed malt or blend of malts to hot water often containing added small quantities of specified mineral salts in solution. Great care is usually taken to control the temperature at which mashing is effected, some of the reasons for this being clear from the following considerations. Firstly, an elevated temperature, usually about 65°C. (150°F.) in the infusion system with which top fermentation brewing is mostly concerned, is necessary if the production of wort is to be completed within a reasonable time, usually about two hours. Of the enzymes α- and β-amylase, which are jointly concerned in the breakdown of starch, β-amylase appears to

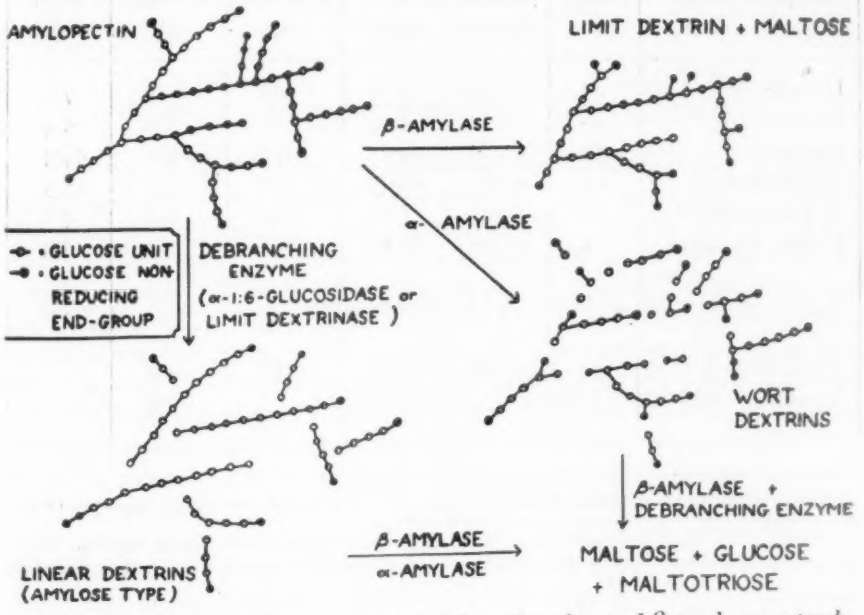


FIGURE 12. *Diagramatic representation of the action of α- and β-amylase on starch*

act by attacking only the non-reducing ends of the chains of glucose units (Figure 12) in such a way that the latter are removed two at a time in the form of maltose. This action, however, seems not to be possible beyond a point of branching. It follows from the picture of starch structure already developed that whereas the molecules of amylose might in this way be broken down almost completely (in fact it is usually far from completely disrupted due possibly to such factors as an inhibitory action of the break-down products on the enzyme itself), the splitting of amylopectin would be much less complete. It would presumably only extend to the point where the side-chains (see Figure 9) already shortened during the malting process were curtailed altogether. Under normal conditions, however, the action of β-amylase is supplemented by that of α-amylase which is able in addition to split those α-1:4-linkages which occur at internal points of the chains of glucose units. In this way new reducing ends are produced which are susceptible to the further attack of β-amylase. Theoretically therefore a fully degraded wort should on this basis consist largely of maltose together with a series of polysaccharides representing the fragments of the branched structures of the original amylopectin molecules. The detailed composition of wort has in recent years been extensively investigated largely by means of paper chromatography[16] and typical results using a colorimetric technique to estimate the individual components are illustrated in Figure 13.

CARBOHYDRATE COMPOSITION OF WORT BY CHROMATOGRAPHIC ANALYSIS

Sugar Fraction	*Vol. of Eluate (ml)*	*Nett Optical Density*	*Hexose Concn.* μg/ml*	*Total Hexose in Eluate μg*	*Total Sugars in Eluate μg*	*% Wort Solids*
Hexose	25	0·132	9·10	228	228	7·2
Sucrose	20	0·131	9·05	181	172	5·4
Maltose	50	0·426	29·42	1471	1397	44·4
Trisaccharide ...	25	0·267	18·42	461	430	13·5
Tetrasaccharide ...	20	0·105	7·24	145	134	4·2
Higher Dextrin ...	25	0·375	25·86	646	582	18·3
Total Carbohydrate	in 0·03	ml† (by	Summation)		2943	93·0
Total Sugar† ...	50	0·306	21·15	1058	998	
Total Carbohydrate	in 0·03	ml (by E	stimation)		2994	

* Nett O.D. × 69—Hexose concentration in μg/ml.

† For each chromatogram 0·03 ml of wort was loaded on the paper whereas in the direct estimation of total sugar only 0·01 ml was used.

NOTE: The wort sample used in this analysis had a specific gravity of 1041·66. Since the papers were loaded with 0·03 ml, this represented total addition of solid material of 3180 μg $\left(\text{(i.e., } \frac{41{\cdot}66}{3{\cdot}93} \frac{0{\cdot}03}{100} \mu g\right)$

FIGURE 13. *Typical carbohydrate composition of wort*

It is notable that while maltose is, as expected, produced in largest amount the quantities of glucose and more particularly of sucrose are greater than could be accounted for directly by the amounts of these sugars present in the original malt. There must be therefore a small amount of invertase action taking place and it is probable that this same action is producing small amounts of higher polysaccharides which are not directly related structurally to the starch from which they stem. G. Harris and I. C. MacWilliam[17] have in fact recently isolated what is probably a glucodifructose from wort and which possibly originates in this way.

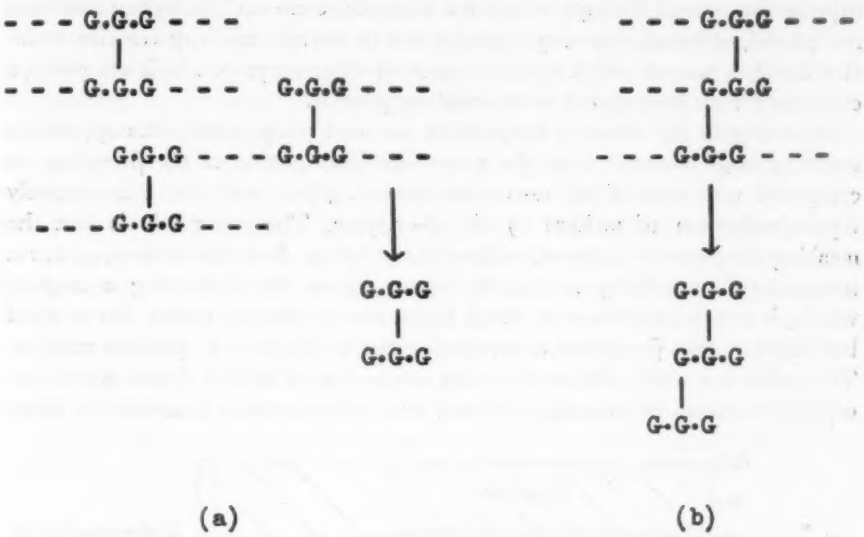


FIGURE 14. *Diagrammatic representation of dextrin formation*

Most of the polysaccharides of wort, nevertheless, may be pictured as derived directly from starch as a result of the occurrence of branched (α–1:6–) linkages. The combined action of α– and β-amylase is able to shorten the side-chains ideally and ultimately as far as a glucose unit adjacent to the glucose molecule actually forming part of the bridge. This is assumed because of the distinction which can be drawn between the linkages concerned in the branched and straight chains. If therefore the cross-linkages in any one chain are separated by at least one additional glucose unit as depicted at Figure 14 (*a*) the resulting dextrin will be a hexasaccharide. On the other hand contiguous branches as depicted at (*b*) will lead to more complex polysaccharides. Actually the dextrins of wort appear to consist of a mixture with the average complexity of a decasaccharide, a finding which is in satisfactory agreement with prediction. From another point of view it will be seen from Figure 14 that, if the process of dextrinization were to lead wholly to the simpler molecules as shown at (*a*), three glucose units out of each chain in the amylopectin would appear as dextrin. As each chain contains on the average about 16 units and as the amylopectin comprises some

eighty per cent of the total starch, the dextrin would account for three-sixteenths of eighty per cent or about 15 per cent of the wort sugars assuming that all the reactions passed to completion. This estimate is, it must be emphasized only a first approximation, particularly as it assumes that the break-down of the lesser component, amylose, is complete, an assumption which is not borne out experimentally. Actually about twenty to thirty per cent of the wort solids consists of dextrins under normal condition, so that the mechanisms so far considered are more than sufficient to account for the observed formation of higher polysaccharides. Nevertheless the existence of an enzyme in malt capable of disrupting the branch linkages which are resistant to α– and β-amylase has been recognized, although any importance it has in normal mashing remains to be elucidated, a remark which applies to several other enzymes which are perhaps concerned with finer details of the mashing process.

Reverting to the effect of temperature on wort composition, this appears to result in large measure from the more rapid inactivation of the β-enzyme as compared with that of the α–enzyme taken together with the comparatively rapid production of maltose by the β–enzyme. The net result is that the mashing temperature is raised, relatively more of the β-enzyme is destroyed. An increasing responsibility is thereby thrown on to the remaining α–amylose which, it is true, continues to break the chains at internal points but is itself less efficient than β-amylase at continuing the break-down to produce maltose. The result is a wort with an increasing proportion of dextrin which is not susceptible to attack by normal yeasts and thus is decreasingly fermentable. Some

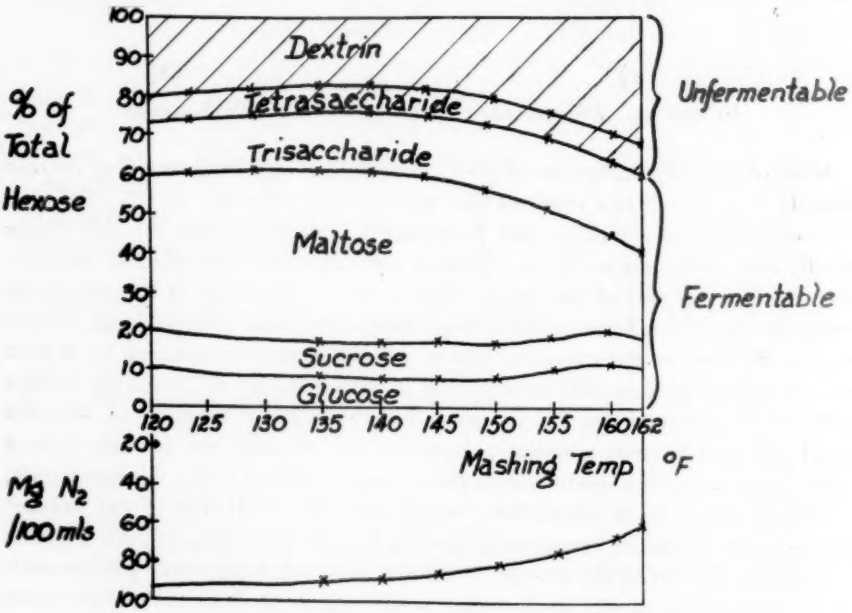


FIGURE 15. *Variation in wort composition with mashing temperature*

recent results in studying the effect of the temperature of mashing on wort composition are summarized in Figure 15. It will be apparent that there is a serious diminution in the yield of fermentable sugars above about 145°F. and indeed the range 145-155°F. is quite critical with regard to wort composition. It may be concluded in fact, taking into account the decreased rate of reaction at lower temperatures, that the normal temperature of mashing at 150°F. represents a satisfactory compromise between the opposing factors involved.

In dwelling on the carbohydrate composition of wort it must not be supposed that the nitrogenous constituents should be neglected. The bulk of the proteolytic activity of green malt is destroyed on kilning but some remains. Although the enzymes concerned appear when isolated to be inactivated somewhat below normal mashing temperatures, they appear to exert *in situ* an appreciable effect on mashing. For instance, about fifty per cent more nitrogenous material can be extracted at a higher temperature than at a low temperature. On the other hand the progressive inactivation of the enzyme is apparent from the lower section of Figure 15. Indeed it might be concluded that one of the disadvantages of mashing at a temperature lower than normal would be the increased amount of nitrogenous material taken into solution with a possibly increased tendency to produce haze in the final beer. Recent results. however, indicate that the haze potential of beers from wort produced at higher temperatures may be greater than from mashes at lower temperatures. One must conclude either that there is some difference in the nature of the nitrogenous materials produced at the higher temperature, or that any relation between haze potential and the amount of nitrogenous material is conditioned by another, so far unelucidated, factor which has an enhanced effect as the temperature of mashing is raised.

HOPS

The hops used in brewing consist of the dried female flowers of the dioecious perennial *Humulus* species. The cultivation of hops is a traditional branch of agriculture, having long been intensively carried on in parts of Germany and in this country, notably in Kent, the West Midlands and to a smaller extent in other parts such as the Surrey-Hampshire border to which William Cobbett referred. The plant is also cultivated in the eastern regions of the United States and more extensively near its western seaboard while hops are now being produced in increasing quantities in New Zealand and South Africa. In this country the permanent sets start into growth in early spring and the rapidly growing vines are given constant attention by way of fertilizers, sprays and stringing or similar support. It is common here to include a number of male plants in each garden, with the result that the final hops are both larger and seeded while on the Continent, and to a varying extent elsewhere, it is usual to exclude male plants so that the hop cones are unfertilized. The time of picking, in England usually around the end of August, is selected to ensure hops of the best brewing quality. Moreover as the mature hops deteriorate very quickly in the fresh green state and as the picking season is short and the equipment generally elaborate, costly and specialized, the organization of the culminating stages of hop production requires extraordinary care and experience.

The quality of the ultimate hops can easily be marred by incorrect curing, which consists in drying under controlled conditions of temperature, air-speed and time, usually in this country in the presence of a small quantity of sulphur dioxide. The latter is commonly introduced by burning sulphur in the oast house, but the direct use of gaseous sulphur dioxide may be preferred in the future. The use of sulphur is favoured in England because of the silky yellow appearance which the hops thus acquire, though actually the bittering power of the hops seems to be thereby diminished under normal conditions. All these conditions affecting hops production have been and are still being intensively studied in this country notably at Wye College, Kent[18]. Added impetus is given to these investigations by the increasing use of machines for picking hops and as well by the need for new varieties of hops which shall be less susceptible to serious plant diseases and at the same time fully acceptable to the brewer.

The study of the hybridization of hop varieties is inseparably linked with the name of Professor E. C. Salmon, of Wye College, who has devoted himself to this work over upwards of half a century. The etiology and other aspects of diseases of the hop plant have come under intensive examination more recently, notably at the East Malling Research Station, and as a result resistance or tolerance to disease, particularly to Verticillium Wilt of hops has emerged as an important if not indeed the first agricultural requirement of any new hybrid hop which may be offered to the brewer. Remarkable progress has been made in the development of promising new hops but their wide acceptance by the brewer is subject to many other considerations. For instance, quite apart from their different agricultural response to fluctuating climatic conditions from season to season, the flavouring characteristics of hops invariably alter, often to very different extents and mostly in the direction of deterioration on storage. Although this deterioration is lessened by storage in the cold as is now becoming common, it remains one of many important considerations to the brewer in his endeavour to maintain an unvarying standard from brew to brew throughout the whole year between the annual harvests. For such reasons scientific appreciation of the part played by hops in brewing has become more necessary than ever before.

In present brewing practice hops fulfil a variety of functions of which the more important are to impart a desirable aroma and a pleasant bitterness to the beer. Hops are thought also to increase the resistance of the beer to bacterial infection to which it is liable as a result of its composition, particularly in respect of its unfermented carbohydrates, nitrogenous components and vitamins. Hops are mostly used by adding them, commonly at the rate of one to one and a quarter lb. per barrel, to the sweet wort before or during boiling for about two hours. On cooling the boiled wort, the separation of the protein sludge so formed is assisted by tannins contributed by the hops while the actual filtration is facilitated by the open spongy consistency of the spent hop cones. The latter consideration is however a purely mechanical one, the importance of which may well recede as centrifugation comes into wider use. Scientific effort has been indeed more particularly directed to the other functions already mentioned.

Wort boiling is an integral part of brewing, for by this means a relatively stable wort is produced chiefly by the inactivation of any enzymes remaining after mashing and by the precipitation of protein. The addition of hops at this stage involves the addition of a variety of nitrogenous compounds, carbohydrates[19] and pectins[20] which pass into the wort, although the amounts involved are probably too small to be significant. In addition, however, a proportion of the essential oil of hops is taken up to give the final beer its aroma, while certain fractions of the resinous constituents of hops account both for the ultimate bitterness and for the slightly antiseptic properties of the beer.

The direct use of hops in wort boiling in open vessels results in at least ninety per cent of the essential oil being lost while much of the residue becomes resinified. Under working conditions this may not be entirely undesirable, for the process to some extent eliminates unwanted aroma and it is believed that the resinous transformation products retain the original fragrance. It is obviously unlikely that this retention is complete, and for such reasons it is not unusual to add part of the total hops used at a late stage during boiling or even to treat the finished beer in cask with a small quantity of hops ('dry hopping'). Frequently hops specially selected for their fragrance are used for this latter purpose. This practice is, of course, only one reflection of the fact that the fragrance of hops of different varieties can vary markedly while the aromatic properties of isolated hop oil are notoriously inconstant because, among other reasons, they deteriorate on storage.

The physical or chemical assessment of the quality of hop oil is at once made difficult by the fact that the oil constitutes only 0.2 to 0.5 per cent of the hop cone. R. G. Wright and F. E. Connery[21] found only an approximate relationship between the 'quality' of the oil as judged by organoleptic tests and the proportion boiling above 190° while J. G. Lambert[22] claimed that the ester index and the hydrocarbon content of the oil have some significance in this connection. It is clearly desirable to know more precisely the individual constituents which are concerned and here the important work of A. C. Chapman[23] may be recalled. He isolated from hop oil four compounds regarded as myrcene, linalool, geraniol and linalool isononoate together with four new compounds named humulene, now regarded as α-caryophyllene, luparone, luparol and luparenol. Luparol appears to be a phenolic ether and luparenol a sesquiterpene alcohol. F. Sörm and his colleagues[24], using chromatography on alumina, obtained a pelargonic but no isononoic ester, methylnonyl-ketone which is probably identical with Chapman's luparone, farnesene (thus obtained for the first time as a naturally occurring compound) and a terpene resembling β-caryophyllene. Obviously the problem is extremely complex and particular interest attaches therefore to the application of two new and refined techniques. The first of these[25] employs chromatoplates which consist of strip chromatograms on an absorbent which has been spread on glass plates. The initial chromatograms are substantially colourless but the positions of various components can be revealed by suitable reagents applied as sprays or by inspection in ultraviolet light. By using alternative solvents, either the hydrocarbons or the oxygenated components of the oil may

be examined selectively. A close examination of typical chromatograms revealed some six hydrocarbons and at least twice as many oxygenated compounds. Moreover, there were considerable differences between the patterns given by hop oils of different origins, histories and qualities. The second technique to be adapted to this problem recently has been gas-liquid chromatography[26].

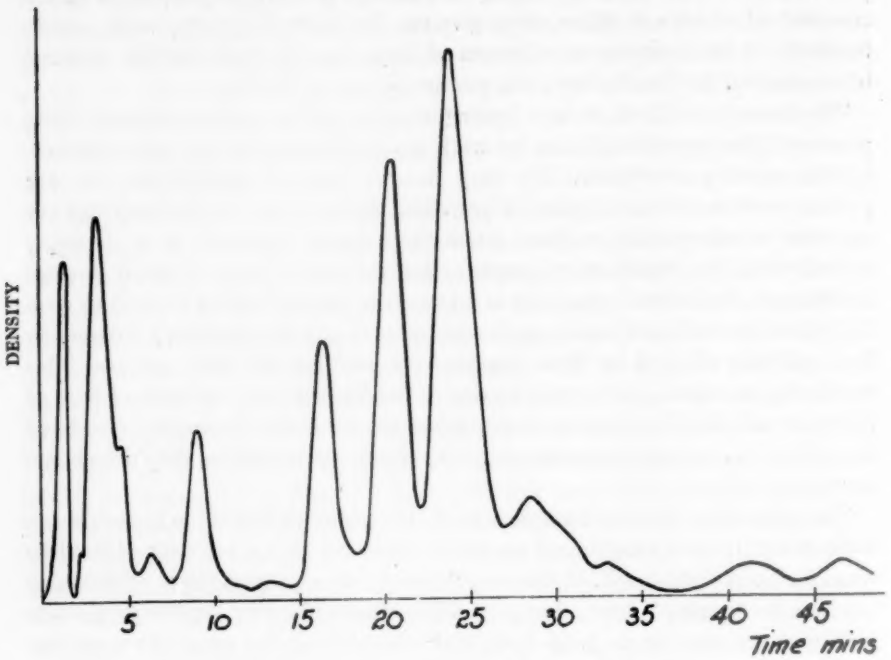


FIGURE 16. *Typical pattern obtained from gas-liquid chromatography of hop oil*

The method consists essentially in passing a minute quantity of the oil in an atmosphere of nitrogen through a long glass column containing an inert filling coated with a selected fraction of a lubricating oil. The latter acts as a stationary phase as in a normal liquid-solid chromatogram and in effect retards each component of the hop oil to a characteristic extent. Consequently when the density of successive fractions of the gas issuing from the tube is examined by a sufficiently delicate means, and the determined density plotted on an arbitrary scale against the fraction number, a complex pattern is obtained (Figure 16). In this pattern each peak corresponds theoretically to an individual compound while the area under the peak affords a measure of the relative quantity of the particular substance present. Inspection reveals the presence of at least 18 compounds, while still other more volatile constituents may well have escaped detection. Certain tentative identifications indicate the presence of myrcene, methylnonylketone, α– and probably β-caryophyllene among others, but it is noteworthy that both gas-liquid chromatography and the chromatoplate

technique indicate the absence of geraniol, contrary to earlier work. Clearly such methods offer promising approaches to the useful assessment of hop oils in terms of chemical composition and ultimately of the desirable qualities of aroma.

Fortunately the situation with regard to relating the bittering and bactericidal properties of hops to individual chemical constituents is more satisfactory, though still by no means completely so. These properties do not belong to the hop cone as a whole, but are restricted to the yellow resinous particles (lupulin) which are found near the bases of the petals or bracts (Figure 17).

FIGURE 17. *Detailed structures of hop cone*

Rubbing a sample of hops between the palms of the hands firstly enables the aroma to be appreciated, but more particularly gives some indication of the total resin content and so with experience is a rough guide to the relative value of different hops.

A long series of investigations has revealed that lupulin may be fractionated in the laboratory according to a scheme which effects the segregation of a large part, if not the whole, of the brewing value of the hop or lupulin into a fraction conventionally termed the α-soft resin. The latter comprises about four to five per cent of an average sample of hops as compared with the β-resin which often accounts for some eight to ten per cent of the weight of the hops.

The nature of the former material was investigated by H. Wieland *et. al.*[27] who isolated a crystalline component, humulone. As a result of degradative studies these authors proposed a structure which despite unusual features of chemical reactivity has required only minor modification over the intervening thirty years[28]. Consequently the formulation given in Figure 19 is now generally accepted as representing the structure of humulone. The compound has been synthesized[29], although the method does not unequivocally establish the correctness of the structural representation. Although humulone itself is both bactericidal

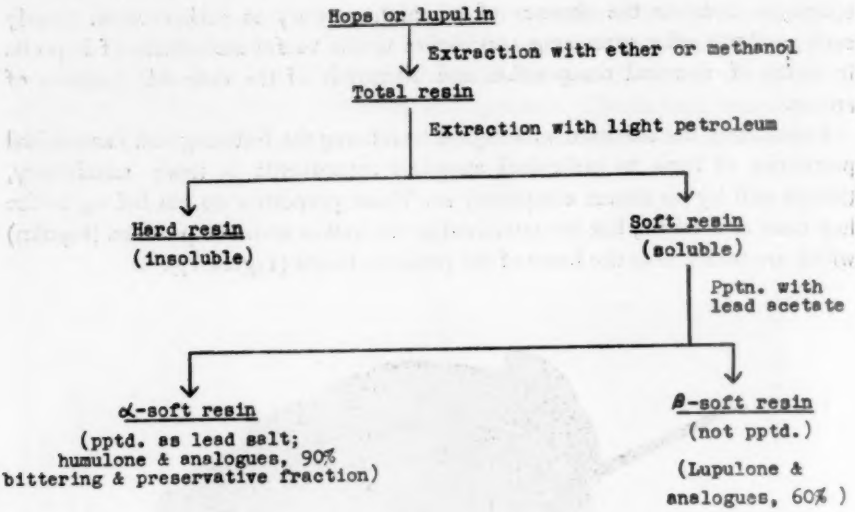


FIGURE 18. *Fractionation of total resin of hops*

and intensely bitter, numerous investigations have shown that there is no immediate connection between the brewing value of a hop and either its content of pure humulone or the yield of lead salt from which the humulone can be isolated.

The first barrier to any such direct connection lies in the fact that on boiling wort with hops the humulone undergoes a complex transformation. Moreover, under normal conditions only about ten per cent of the humulone added as hops passes ultimately into the fermented wort, the remainder being resinified or carried down with the sludge. Unfortunately precise knowledge of the nature of the ultimate bittering and preservative principles derived from humulone is still lacking. For instance, while bitter transformation products have in several laboratories been obtained by treating humulone under various alkaline conditions, it is known that such products are complex mixtures and it is obviously possible that they differ considerably from those formed during brewing where to mention only one likely factor, the material is subjected to slightly acid conditions. Further it is possible, and indeed there is evidence, that bitter transformation products can occur preformed in the hop when their contribution to the bittering of beer would not be reflected in the quantity of α-soft resin originally present.

The second obstacle to any firm link between bittering and bactericidal power on the one hand, and α-soft resin or humulone content on the other, lies in the fact that within the past few years examination of typical α-soft resins, by means of counter-current distribution in the first instance has shown that they are surprisingly heterogeneous[30], and that sometimes humulone may not even constitute the major component. The method consists essentially in partially

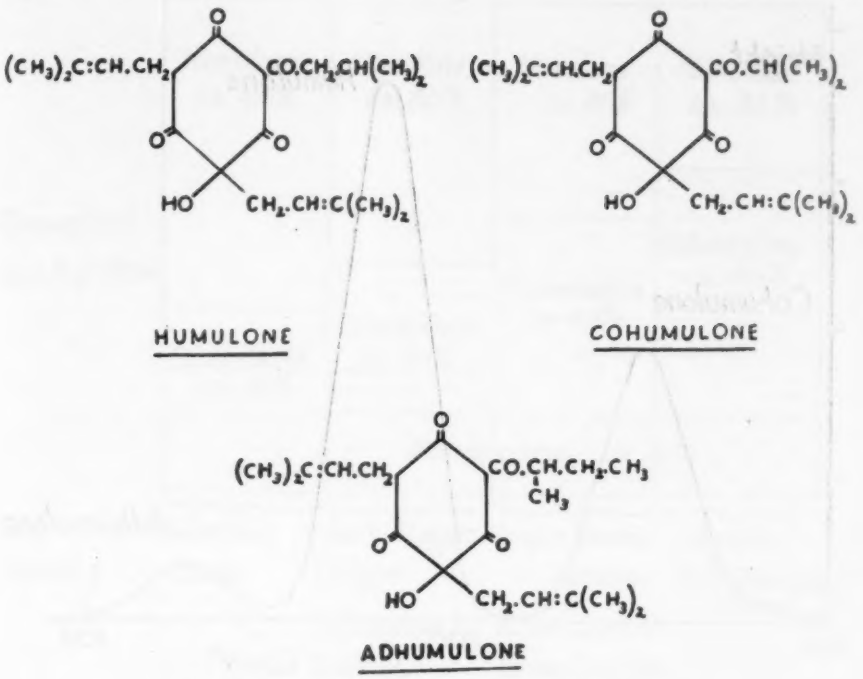


FIGURE 19. *Structure of humulone and its congeners*

extracting a sample of the resin in a solvent such as light petroleum, with a selected aqueous buffer solution. The tube in which the extraction is carried out is arranged so that the partially spent light petroleum solution is transferred to a second tube containing fresh aqueous extractant. Meanwhile, the first tube is charged with a fresh batch of light petroleum. On repeating the process the original light petroleum solution is transferred to a third tube and ultimately after, say, a hundred transfers the initial sample of resin is distributed over a hundred tubes. The long series of complex operations can be carried out completely automatically. If now the amount of resin in each tube be estimated, as can conveniently be done by ultra-violet spectroscopy, the distribution along the series of tubes can be expected to be very uneven. Should the resin contain only one extractable component then a graph relating concentration to the tube number will be symmetrical. Indeed the position of the apex of the peak with reference to the tube number can be predicted from the independently determined distributed coefficient between the solvents employed. If, however, more than one compound be concerned, each will move along the series of tubes at a characteristic rate, so that ultimately the appropriate graph will show a series of peaks. In practice the degree of symmetry of the peaks is a guide to the extent of separation achieved while the area under each peak provides a measure of the relative abundance of the compound concerned. A typical soft resin examined in this way (Figure 20) is seen to contain three main components of which one is, of course, humulone.

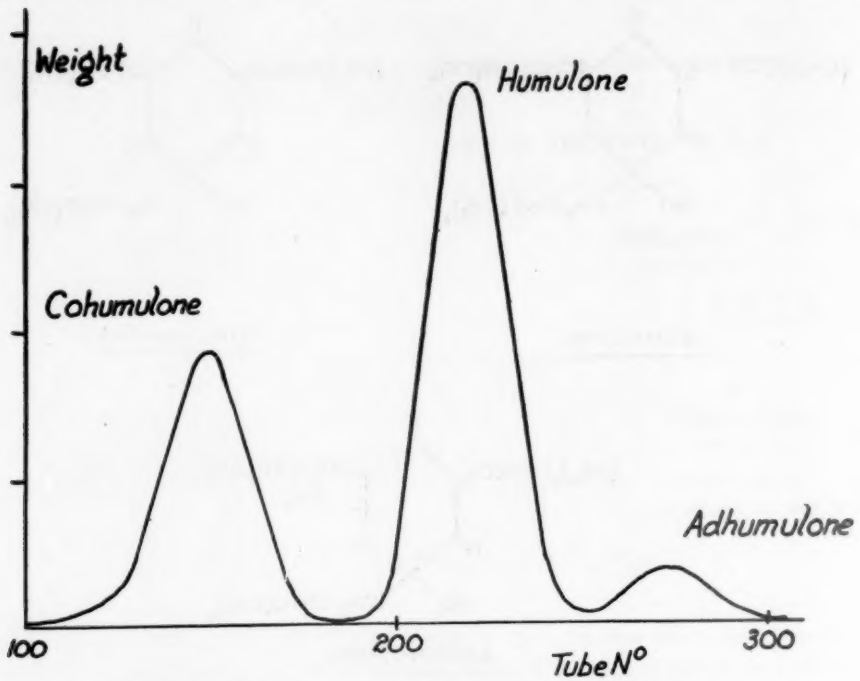


FIGURE 20. *Countercurrent distribution of a typical α-soft resin of hops containing humulone, cohumulone and adhumulone*

The direct isolation of these from the contents of appropriate tubes has revealed that they are in fact all rather similar in chemical, bittering and bactericidal properties. The two new components, cohumulone and adhumulone are indeed a lower homologue and an isomeride of humulone respectively (see Figure 19), the three components differing only in their acyl side-chains. These facts would not concern the brewer if either the brewing values of the individual components were equal or if alternatively the compounds occurred in constant proportions. Actually these conditions are probably immaterial with reference to adhumulone, which mostly constitutes a constant proportion of at most about 15 per cent of the α-soft resin fraction. They apply with considerable force, however, to the relative proportions of humulone and cohumulone. Each of the three components undergoes a so-far ill-characterized transformation in boiling wort as was related above with reference to humulone; the crude 'iso' materials isolated from beer have indistinguishable bittering powers. However, the efficiency with which the original resin fractions are converted into the ultimate bittering substances is substantially different, with the result that a hop with a high proportion of cohumulone confers on a beer a harsher, more bitter flavour than one with less cohumulone even when the two are used at equivalent rates as judged by their α-soft resin contents.

Figure 21 summarizes the compositions of the α-soft resins of a wide variety

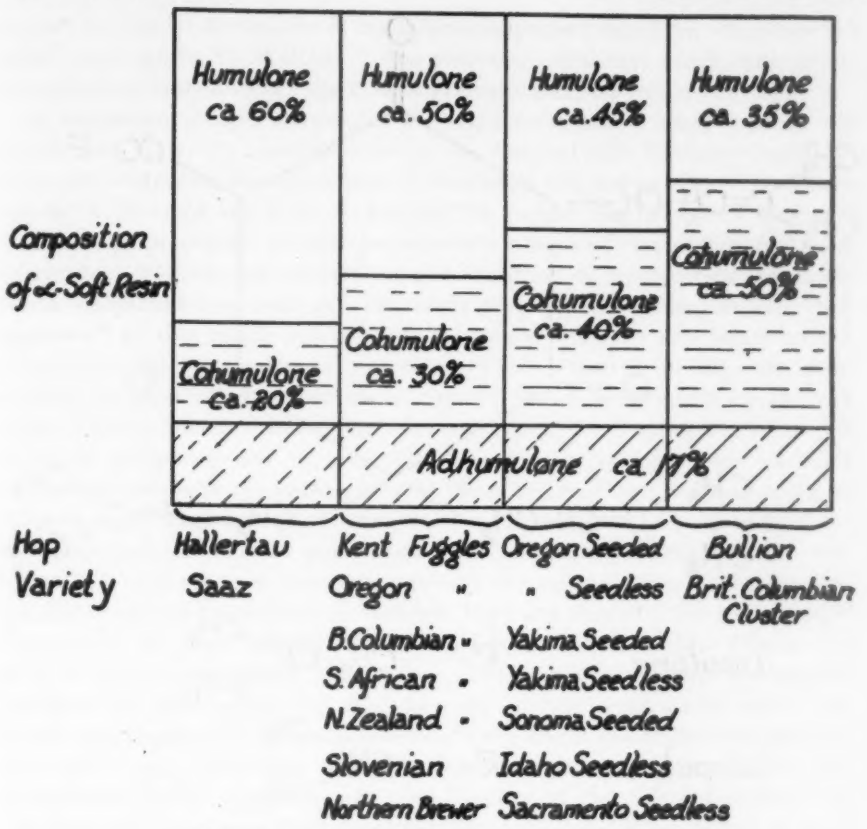


FIGURE 21. *Composition of α-soft resins of hops*

of hops as determined in the first instance by counter-current distribution[31]. These hops fall into four groups in which the α-soft resins contain about twenty, thirty, forty and fifty per cent respectively of cohumulone and it is specially interesting to find that in conformity with what has already been said this arrangement results in the more delicately flavoured hops appearing together at the left of the diagram, with those usually regarded as less desirable coming towards the right.

It has already been mentioned that many of the above analyses were carried out by means of counter-current distribution but the method is obviously extremely time-consuming. More recently G. A. Howard and A. R. Tatchell[32] have evolved a more convenient procedure suitable for routine use. They oxidize the α-soft resin so that the different side-chains of the individual components each affords its characteristic fatty acid. Analysis of the mixture of fatty acid by means of gas-phase chromatography thus rapidly gives the composition of the original resin.

It should be mentioned that the above compositions do not reflect the total range possible. For example a wild American hop has been found, in which

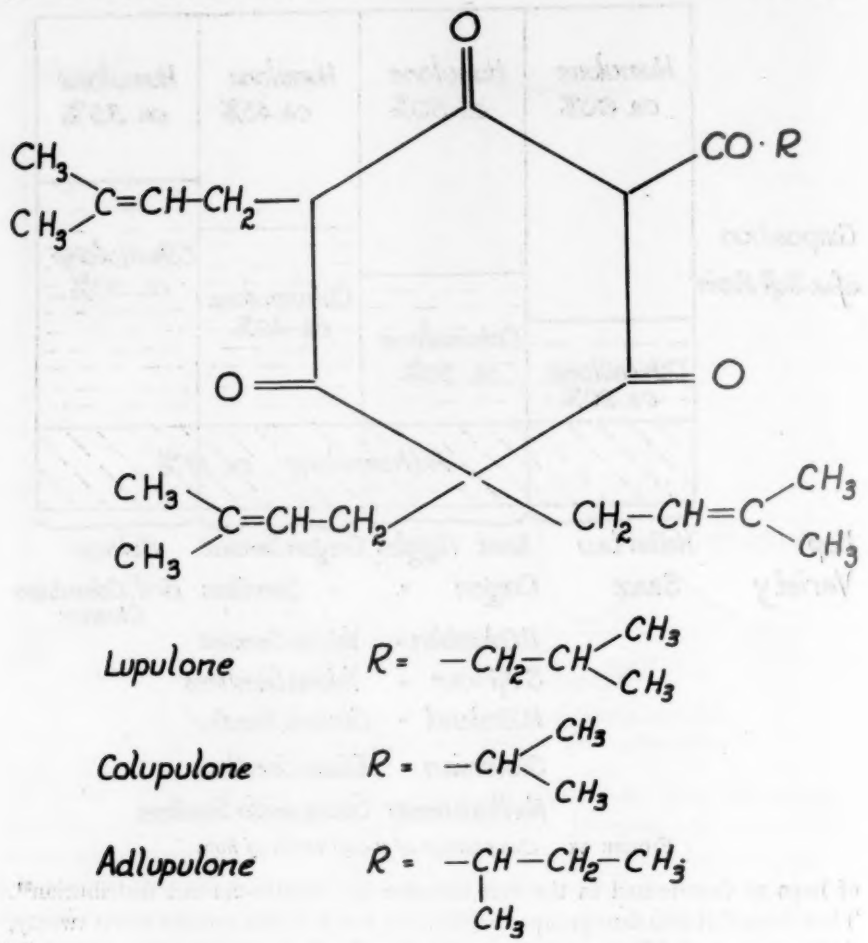


FIGURE 22. *Structure of lupulone and its congeners*

colupulone comprises no less than eighty per cent of the α-soft resin. As many of the varieties shown are descended from hybrids between this and Kent Fuggles, it appears that successive crossings reduce the percentage which thus seems subject to genetic control. On the other hand, it has been found[33] that the composition of the α-soft resin alters markedly as the hop cone approaches maturity. The above compositions therefore refer to hops picked according to the dictates of experience in the hop garden and this suggests the advisability of added caution in assessing the value of new varieties of which corresponding experience must inevitably be lacking.

The β-soft resin is both bitter and bactericidal, but under normal brewing conditions makes very little, if any, contribution in these respects to beer. Nevertheless its composition is of considerable scientific interest, especially as this portion of the hop resin represents a substantial potential additional

source of value to the brewer. A crystalline component, lupulone, was examined many years ago by W. Wöllmer[34], who proposed a structure which, with minor modifications embodied in Figure 22, is generally accepted to-day[35].

A succession of later investigators studied a crystalline component of the β-soft resin under the impression that it was identical with Wöllmer's lupulone. However, increasing discrepancies in the analytical data and in other observations led G. A. Howard and J. R. A. Pollock[36] to suggest that, despite a deceptive similarity in properties, two compounds were concerned and that in fact most of the later workers had handled a lower isomeride of lupulone for which the name colupulone was proposed. All doubt that this is the case has now been removed[37] so that taking into account the synthesis of the two compounds[38], colupulone has been assigned a structure which shows it to have the same relation to lupulone as cohumulone does to humulone (Figures 19 and 22). Both counter-current distribution, after subjecting the β-soft resin to certain chemical transformations to overcome difficulties in working with the resin itself, and gas-phase chromatography of the mixture of fatty acids obtained as in the case of the α-soft resin, afford clear evidence of the heterogeneity of the β-material and at the same time considerable insight into its composition. The remarkable fact emerges that both lupulone and colupulone are constant congeners in varying proportions in different hops and that they are accompanied by at least one more compound. The latter is termed adlupulone (Figure 22) as it is the counterpart of adhumulone. There is thus complete parallelism between the two resins, and moreover the limited quantitative data[39] (see Figure 23), suggest that the compositions of the α- and β-soft resins from different hops may vary somewhat similarly. The percentages of colupulone and cohumulone in the respective soft resin fractions of the different varieties are not the same, but it seems significant that they are related in a constant manner, so that the percentage figure for cohumulone seems always to be about two-thirds of the corresponding figure for colupulone.

Hop variety	% *Colupulone in β—soft resin*
Hallertau	37
Northern Brewer	45
Kent Fuggles	49
N. Zealand Fuggles	64
Oregon Seedless	60
Bullion	70

FIGURE 23. *Composition of the β-resin of different varieties of hops*

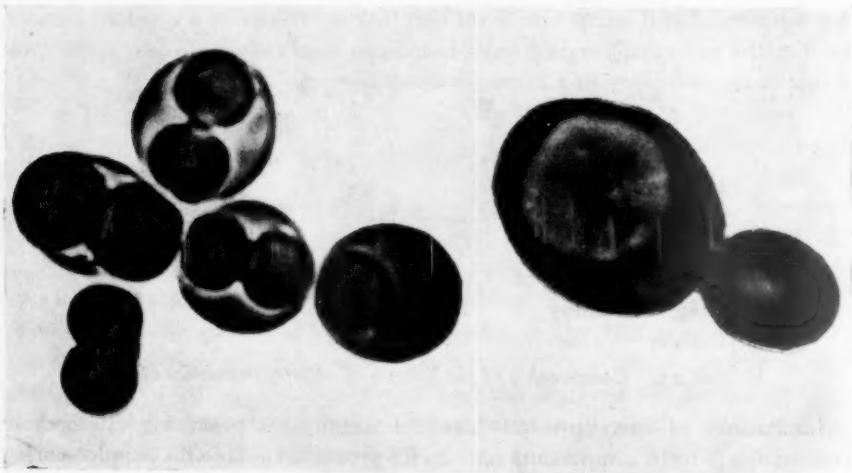
Limitations of space preclude detailed mention of promising attempts to convert the β-resin components into useful products suitable for supplementing the α-compounds in brewing, but it will be clear that chemical research has already played a large part in the detailed assessment of hop varieties and it seems likely that many of the above developments will make a still larger contribution in the course of time.

LECTURE III

Monday, 5th December, 1955

FERMENTATION: GENERAL CONSIDERATIONS

While the details of the fermentation of wort vary considerably, the essential outlines are common to practically all systems of brewing. Thus in top fermentations there is for a few hours after adding the yeast very little change, either in the yeast population or superficially in the composition of the medium. This lag phase is presumably a period in which the cells adapt themselves to the medium in which they are placed. One can postulate changes in the nature of the surface of the yeast cell, the development of the mechanism, hitherto latent, of reproduction, and the production of enzymes necessary to effect the many individual reactions associated with fermentation and one can envisage many other changes brought into action at this stage. They can perhaps all be summarized by regarding this period as one in which the cells become efficiently organized to effect the fermentation. After five to eight hours the cells begin to multiply vegetatively very rapidly (Figure 24*b*), until after about 24 hours the population with regard to live yeast reaches almost its limit. Yeasts are frequently able to multiply by sporing also (Figure 24*a*). This process is naturally unwelcome in the brewery, as it may lead to strains of yeast differing in properties from the original culture, whereas the strain characteristics remain unchanged when the yeast multiplies vegetatively except in the relatively rare occurrence of mutation. The production of spores is, on the other hand, essential to any process of hybridization of yeasts whereby desirable properties of separate strains can



(*a*) (*b*)

FIGURE 24. *Multiplication of yeast cells;*
(a) *by sporing* (b) *vegetatively*

perhaps be stably united in a single yeast. Unfortunately, the production of spores is fickle with many yeasts and indeed with some does not appear to occur at all. Considerable interest attaches, therefore, to the demonstration that the process is associated with the production of a substance or substances which can stimulate formation of spores by other cells[40], but adequate experimental control over the whole phenomenon has still to be attained. The so-called log phase of growth is seemingly characterized by a predominantly aerobic metabolism, as the curve showing its progress with time is practically the mirror image of that depicting the drop in pH (Figure 25), a change which is characteristic of the purely oxidative

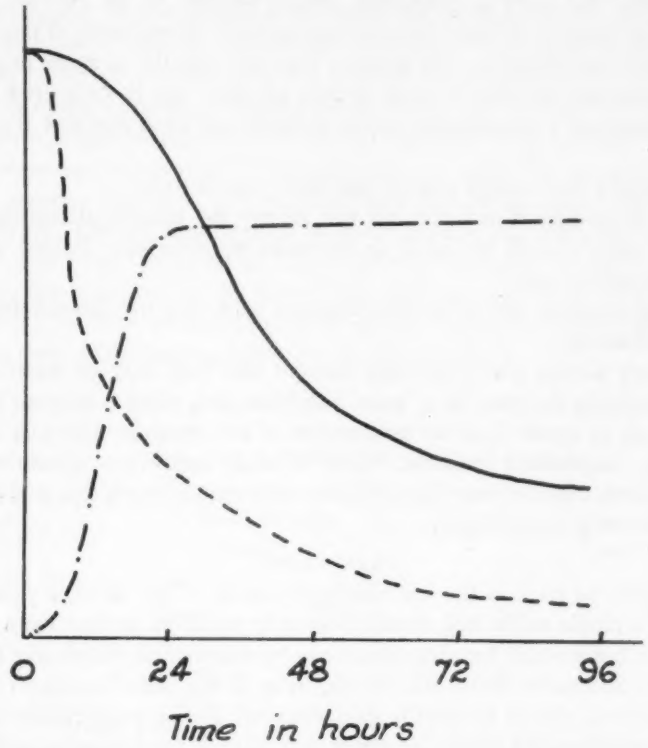


FIGURE 25. *Diagrammatic representation of growth (-.-.-.); change in pH (- - -) and change in gravity (—) during a typical fermentation*

break-down of sugar. As was mentioned earlier, the normal five to seven-fold-increase in yeast actually accounts for about one to two per cent of the wort solids, so that the gravity of the wort changes only to a very small extent over this initial period. After about 24 hours, however, an increasing proportion of the usage of wort solids is to be ascribed not to multiplication of yeast cells but to fermentation by the existing yeast population, with the result that the gravity falls steadily over the second day and then more slowly until the attenuation reaches its practical limit.

For a variety of reasons fermentations in actual practice may show substantial deviations from the typical behaviour. For instance, the log period may be unduly prolonged, so that in one exceptional yeast, maintained at the Brewing Industry Research Foundation, some two to three days are required before the internal organization of the yeast cells is sufficiently geared to their multiplication preparatory to fermentation. In other cases the yeast may flocculate unduly early and as important stages of the fermentation are effected within the cells themselves this can result in much of the fermentable material escaping breakdown, that is to say, in incomplete attenuation. Still again, the yeast in question may ferment the wort incompletely, either because of an over-all enzymic deficiency or because of inability to ferment specific constituents of the medium. Taking such considerations into account together with the obvious requirement that, for practical brewing, a yeast should not give rise to undesired flavours, three fundamental requirements may be noted in any yeast destined for brewery use:

1. It should have a high rate of vegetative reproduction;
2. the fermentation velocity and the affinity for individual constituents of the wort should be such as to make possible the desired degree of attenuation; and
3. the flocculence should be of a character such that the desired attenuation is achieved.

Subsidiary factors also come into account and here may be mentioned the need to maintain the yeast in a 'pure' condition and, when a mixture of strains is employed, to ensure that the proportions of the component strains are as far as possible maintained constant. Some of these subsidiary questions are so important with regard to meeting the three main requirements that it is necessary to consider them more closely.

'PURE' YEASTS

In order to be pure in the microbiological sense of the word, a yeast should consist of a single strain and should obviously therefore be free from bacteria. In practice few normal brewing yeasts can be maintained completely free from bacteria in successive fermentations as, even if the yeast inoculum be satisfactory, bacteria almost invariably gain entry *via* the brewing vessels or liquor. The practical objective in this respect is, accordingly, to exercise microbiological control so that the bacterial contamination does not exceed a selected limit. Opinion varies widely with regard to what is tolerable contamination, for while on the one hand a proportion as low as five bacterial cells per million yeast cells is ordinarily difficult to achieve, brewery yeasts containing five per cent of contaminating bacterial organisms are sometimes encountered, usually because of the difficulties so arising. The actual proportions can be determined either by direct plate counts, or by counts which have been facilitated as a result of suppressing the growth of the yeast by incorporating a specific antifungal agent such as actidione in the medium.

Apart from what may be termed normal attention to brewery hygiene, it is attractive to consider the use of antibiotics which shall be selective so as to

eliminate generally any bacterial flora from yeast cultures, but preferably not specific as to the latter. To this end much attention has been given to the polypeptide antibiotic *polymyxin*. No less than five materials of this name are known, of which polymyxin B has come into prominence in control tests in the brewery[41]. It has been claimed that polymyxin in conjunction with another substance, thiolutin, exerts a high degree of control over bacterial infections in bottled beer. In view of the fact that the common 'brewing' bacteria include both Gram-positive as well as Gram-negative types and that polymyxin is comparatively ineffective against Gram-positive bacteria, this claim is of considerable interest. As the use of polymyxin in this way would however contravene the existing law of this country greater immediate importance attaches to the possibility of using the antibiotic to free yeasts from unwanted bacteria. A detailed study of this kind using yeasts deliberately infected with various brewing bacteria drawn from about seventy strains actually isolated from brewing yeasts and spoiled beers has shown that infections due to authentic *Flavobacterium proteus*, a Gram-negative organism, can be practically completely controlled by means of polymyxin. The effect is exerted by incorporating the antibiotic into a wort suspension of yeast for 24 to 48 hours and experimentally it can be observed by adding the antibiotic to the fermenting medium. Quantities of 0·005/g./ml. are effective in beer while amounts of several thousand times this quantity appear to have no deleterious action on the yeast itself.

Time of sample hours after pitching	*Number of bacteria/ml. of wort*			
	a) *Flavobacterium proteus*		b) *Acetobacter oxydans*	
	Control	*Test*	*Control*	*Test*
2	50×10^4	$14{\cdot}4 \times 10^3$	$23{\cdot}5 \times 10^4$	13×10^4
24	$87{\cdot}5 \times 10^4$	38	$16{\cdot}9 \times 10^4$	14×10^4
48	$10{\cdot}9 \times 10^2$	0	48×10	$44{\cdot}5 \times 10$
72	$27{\cdot}5 \times 10^2$	0	$10{\cdot}9 \times 10^2$	$77{\cdot}5 \times 10$
Yeast skimmings at 72	$23{\cdot}5 \times 10^4$/gm.	3/gm.	$34{\cdot}5 \times 10^3$/gm.	16×10^2/gm.

FIGURE 26. *Use of antibiotics in controlling bacteria in brewing yeast*
Effect of polymyxin on yeasts infected with (a) *Flavobacterium proteus* (b) *Acetobacter oxydans in hopped wort. Scale of fermentation = 16 litre*

Unfortunately, authentic *F. proteus* is probably less common as a yeast contaminant than is often supposed and in any event is usually regarded as harmless. Indeed it is sometimes considered as of little more importance than as an indication of the probable presence of more harmful bacteria. The mere suppression of this one organism may therefore be profitless, as apart from the invariable persistence of resistant strains it may leave other harmful bacteria unaffected. Indeed a closer study has shown that Gram-negative *Acetobacter* infections in yeast are considerably resistant to the action of polymyxin while Gram-positive *Lactobacilli* are practically unaffected at all reasonable concentrations. These limitations are implicit in the results of a still more recent study[43], and lead naturally to attempts to use mixtures of antibiotics to cover adequately the spectrum of bacterial infections normally encountered.

Quite apart from bacterial contaminants, the possibility of the presence of wild yeasts in brewing has also to be taken into account. The brewer normally employs only strains of *Saccharomyces cerevisiae* in top fermentations or *S. carlsbergensis* in bottom fermentations. Many other *Saccharomyces* species and numerous representatives of other Genera including *Pichia*, *Candida*, *Kloeckera*, *Hanseniaspora*, *Torulopsis* and *Rhodotorula* species are met with varying frequency[43]. It is curious that as was first pointed out by L. Walters and M. R. Thistleton[44], wild yeasts appear to be distinguished from the brewer's culture yeasts by their ability to grow on a medium containing the aminoacid lysine as the sole source of nitrogen. Extension and amplification of this observation by E. O. Morris (unpublished) provides a method whereby the wild yeast population of a yeast culture may be fairly easily ascertained. The individual yeasts are of course identified by standard mycological techniques, but in some instances the classical methods have recently been considerably shortened. For instance the ability of yeasts to ferment to varying extents a range of sugars forms an integral part of the identification. This ability is conveniently ascertained by charging a series of glass capillary tubes with a suspension of the yeast and a solution of the sugar under examination. After plugging the tubes with plasticine and keeping them for periods varying from twenty minutes to three hours, active fermentation is indicated by movement in the liquid level and ultimately by expulsion of the plug due to the pressure of carbon dioxide (Figure 27).

The whole test is so rapid as to dispense with the need for rigidly aseptic conditions[45].

THE RECOGNITION OF INDIVIDUAL STRAINS OF BREWING YEAST

Even supposing that a particular brewing yeast has been obtained completely free from bacteria and from wild yeasts it is not unlikely that it still consists of a mixture of different strains of the same species. The preference for certain yeasts by brewers and the fact that it has been worth while to make some of them commercially available is sufficient to show that their influence on the course of brewing and on the final beer is regarded as considerable.

Various attempts have been made, though only in quite recent years, to classify strains of brewing yeast according to their growth requirements and similar

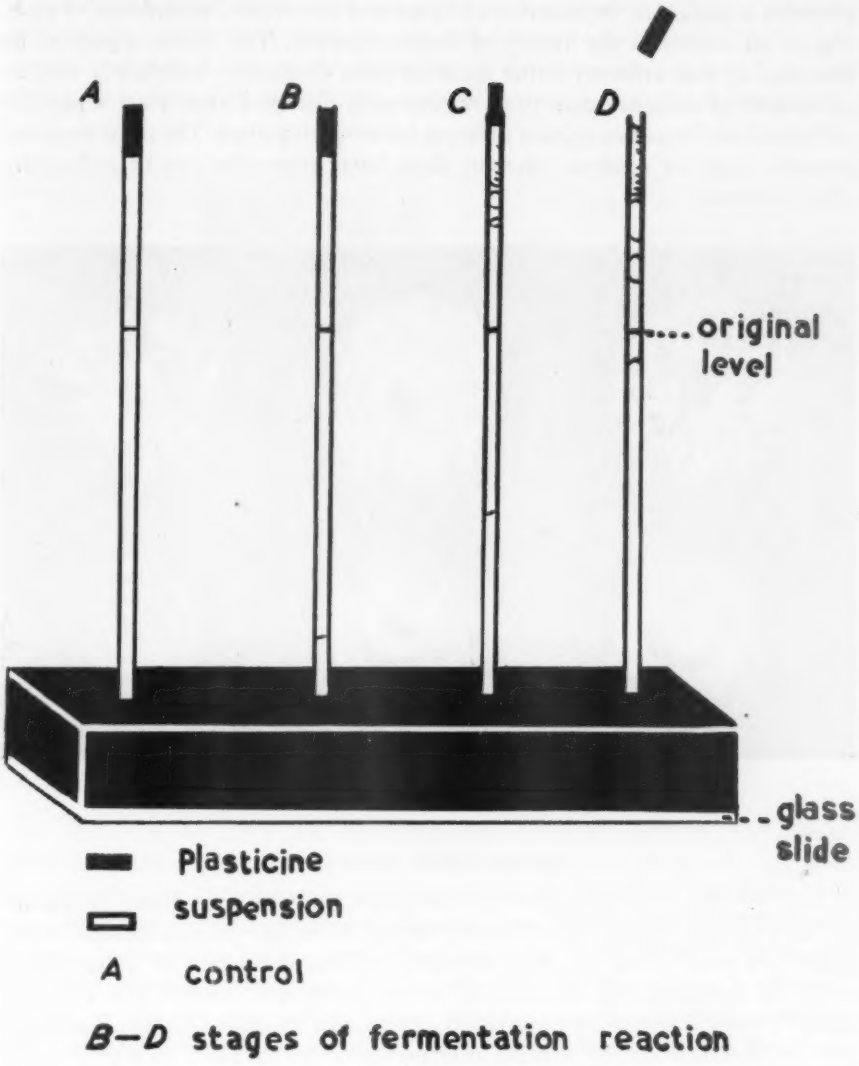


FIGURE 27. *Microfermentation tests on yeasts*

characteristics[46]. With particular regard to yeasts used in British breweries it was, however, first important to devise methods by which the component strains of a mixture might be segregated and then to evolve means by which their more salient features might be compared. Perhaps the best means of achieving the first object is to isolate a considerable number, say a hundred single cells from an appropriate culture and so prepare a hundred separate sub-cultures which are subsequently grown as giant colonies on wort-gelatin[47]. Inspection of the colonies then enables them to be grouped according to their form and so

provides a picture of the number of forms and the relative abundance of each. Figure 28 illustrates the variety of forms observed. The forms appear to be invariant in that different forms maintain their distinction indefinitely and an abundance of evidence goes to show that such distinct forms when separately cultivated can be shown to have different brewing properties. The need therefore presents itself for methods whereby these latter properties can be sufficiently characterized.

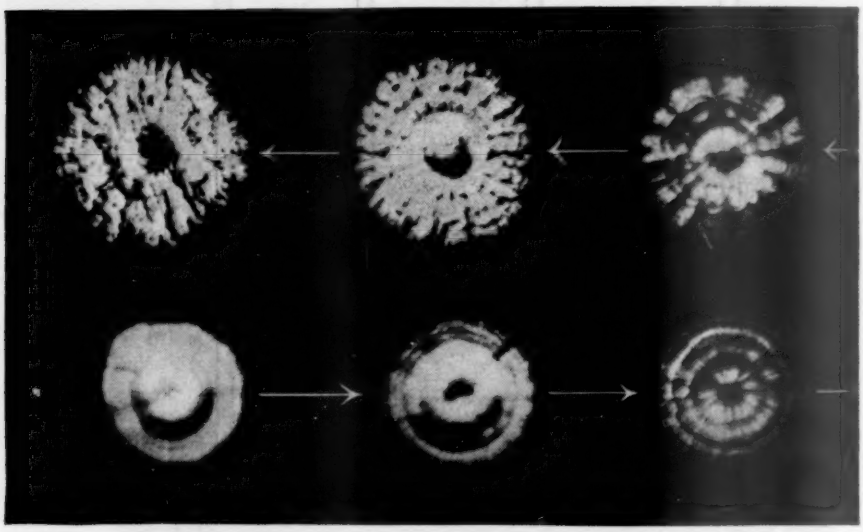


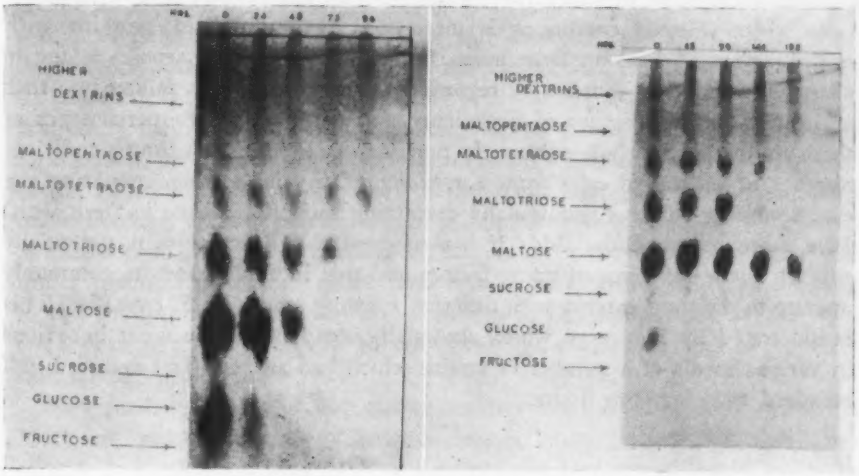
FIGURE 28. *Giant colony forms of a brewing yeast*

FERMENTATION VELOCITY

The velocity of fermentation by a particular yeast has been found to depend on the experimental wort conditions[48], as well as on the nitrogen content of the yeast[49]. A mixed brewing yeast naturally exhibits a velocity of fermentation which is approximately the average of the constituent strains and a recent study[50], using a special fermentometer, shows that the individual strains are in fact characterized by the amount of fermentation taking place in a given time taken into conjunction with the form of the fermentation-time curve.

The actual fermentation in wort depends on the affinity of the different strains for the individual carbohydrates present and there is little doubt that this affinity varies with the strain. The variation is exaggerated when different species are compared, as will be seen from the comparative chromatograms shown in Figure 29. The importance of this behaviour becomes clear when it is realized that a reluctance to ferment maltotriose, for example, is almost certain to lead to slow and incomplete fermentation as this sugar commonly accounts for about 15 per cent of the total wort carbohydrates. Such difference in behaviour has an enzymic background and it is interesting to note that in certain instances

at least the difference seems to be due not to the formation of additional enzymes with specific activity in the case of yeasts which have a wider affinity for the wort sugars; rather it is due to the formation of different enzymes which contain within themselves different abilities to attack the range of sugars concerned[51].



(*a*) (*b*)

FIGURE 29. *Fermentation of brewers' wort with* (a) *Saccharomyces cerevisiae and* (b) *Brettanomyces bruxellensis*

YEAST FLOCCULENCE

It will be appreciated that the flocculation characteristics of yeast are of paramount importance to the brewer. If, after pitching, the yeast cells aggregate too soon so that the clumps separate either at the bottom of the fermented wort or at the surface as is characteristic of most British systems of brewing, fermentation becomes inconveniently slow so that the desired limit of attenuation may be practically unattainable. On the other hand if flocculation is seriously delayed or fails to occur, difficulties are experienced in satisfactorily removing the yeast by any simple system of skimming it off or running off the beer from the yeast head, with the result that ultimately a bright beer cannot be easily obtained. As the flocculence of brewing yeasts, using the word initially to mean the general phenomenon, seems in practice to be variable, its adequate measurement and eventual precise control are desirable objectives.

Nevertheless, it should perhaps first be pointed out that any study of this phenomenon can easily assume to some extent a subjective nature. Obviously flocculation is primarily concerned with the formation of macroscopic aggregates of cells, but it is often a matter of individual opinion as to how large such aggregates have to be before they can be regarded as 'flocs'. Then again, some yeasts undergo incomplete cell division in such a way that chains or branching

clusters of cells are formed instead of completely independent cells. It would obviously be misleading to compare this behaviour directly with the progressive aggregation of initially isolated cells although, as will be seen later, it can be an incidental factor. The reduction of the overall flocculent behaviour of a yeast to numerical terms offers comparatively little difficulty. For instance, Burns[52] described a method in which the volume of yeast sedimenting after a standard time under selected conditions is measured. Again Gilliland, working with mixed strains of different flocculence, found that for many purposes adequate characterization was afforded by regarding the yeast strains as falling into four arbitrarily defined degrees of flocculence and determining the percentages of these strains in the whole culture. In practice this involves selecting a sufficient number of individual cells from a representative sample, then preparing the corresponding cultures and visually examining their flocculence in fresh wort. In a more recent study, Eddy [53] has revealed that flocculation is potentially subject to the influence of many factors and that individual factors commonly operate to different extents with different brewing strains of *S. cerevisiae*. This is illustrated by Figure 30 which shows the comparative flocculent behaviour in various media of a number of strains which had all been first cultured in a standard wort for sixty hours.

Strain	*Synthetic medium*	0·001*N*-*HCl*	0·001*N*-*HCl* + Ca^{++}	*Standard buffer* + 10% of *maltose*	*Standard buffer* + 10% of *mannose*
1	+	±	±	±	±
2	+	±	±	±	±
3	+	±	±	±	±
4	+	—	+	—	—
5	+	—	+	—	—
6	+	—	+	—	—
7	+	—	+	—	—
8	+	—	+	—	—
9	+	—	+	+	—
10	+	+	+	+	+
11	+	+	+	+	+
12	—	—	—	—	—

FIGURE 30. *Varying flocculation behaviour of a series of 12 yeasts*

It will be seen that the flocculative response of the series of strains to a number of different media is irregular. More extended observations have brought to

light other factors which may operate from strain to strain so that it is possible to regard any individual brewing yeast as characterized by the pattern of its flocculative responses to a variety of influences. The different patterns corresponding to closely related strains of yeast, which may in practice coexist in a single brewing culture, go far to reconcile earlier apparently conflicting views on the importance of individual factors. For instance, whereas acidity, calcium content[54], polysaccharide constituents[55] and proteins[56] are among the various factors which have been considered in this connection, it is now clearly not to be expected that they will all invariably prove of importance. Consequently, it is unlikely that irregularities in flocculence among a number of breweries could be inevitably traced to a single circumstance. On the other hand the observation of the varied flocculation responses of the yeasts concerned to various factors under artificial conditions must afford a delicate representation of flocculence. It seems probable that any undesired alteration of the flocculence of a yeast under practical conditions could thus readily be ascribed either to a change in the environment, that is to say, a change in the composition of the initial wort, or to an innate change in the yeast itself and so the avoidance of the alteration might be facilitated.

Two further aspects of flocculation merit special mention here. Firstly, it has been found that in many instances yeasts when pitched into fresh wort remain in suspension until at least a substantial measure of fermentation has been effected. Clearly either fresh wort exerts a deflocculating action in such cases, or fermented wort contains constituents which actively favour flocculation. In fact there is good evidence that both these effects can play a part in controlling flocculation. Thus A. A. Eddy[57] has observed that in certain cases each of the major fermentation products, carbon dioxide and ethyl alcohol, can assist flocculating action. On the other hand, wort sugars usually assist in maintaining the yeast in a dispersed condition in which case the effect is reduced and eventually disappears as the sugars are depleted by fermentations[58]. This action is quantitatively characteristic both as regards the effect of different sugars on the

Strain	*Mannose*	*Maltose*	*Sucrose*	*Glucose*	*Fructose*
I	3·7	5·2	9·2	>12	>20
II	2·1	2·5	5·2	6·6	>20
III	3·1	4·6	8·2	9·2	>20
IV	2·4	3·3	7·3	9·0	>20
V	—	3·6	6·8	7·6	>20

FIGURE 31. *Percentage (w/v) of various sugars just dispersing a number of flocculent yeasts under standardized conditions*

same yeast and also as regards the response of different brewing yeasts to the same sugar. It is clearly demonstrated by measuring the amount of the selected sugar which it is necessary to add to a flocculent culture under standardized conditions, so as just to disperse the cells (Figure 31). Inspection shows that there is indeed a general parallel between the effectiveness of the series of sugars, including mannose (which, incidentally, is not a constituent or ordinary wort), as they act on the successive yeast strains. These facts take on, however, another light in view of the second aspect which deserves particular mention.

A great many observations, under conditions such as have been outlined, have made it apparent that it is necessary to draw a clear distinction between the ability of a yeast to flocculate and the expression of that ability. A brewing yeast freshly pitched into wort mostly remains in suspension, but this is not solely due to the deflocculating wort sugars for, even when examined under varied laboratory conditions, it fails to exhibit any ability to flocculate. Later it can happen that this ability appears in the yeast even though the organism remains in suspension, for instance because of its being maintained in a dispersed condition by residual sugars. On the other hand it sometimes happens that a particular yeast remains in suspension under a wide variety of experimental conditions. In such cases it must be concluded that the yeast is not merely being prevented from expressing an ability to flocculate, but that it is actually lacking this ability. The reality of this ability to flocculate is convincingly demonstrated by maintaining samples of yeast in various media and examining their flocculation under specified test conditions from time to time. It is then observed that so long as metabolism is possible, that is to say, in presence of glucose, even though the media may be such that no growth (cell division) takes place, most strains of *S. cerevisiae* clearly undergo a change in the sense that while the young cells lack any ability to flocculate the more mature metabolizing cells soon acquire the ability. The circumstances can therefore be represented diagrammatically by Figure 32.

The non-flocculable cells (here represented by an unshaded circle) remain dispersed both in a non-flocculent environment and also in one which may towards most yeasts have become 'flocculent' usually as a result of fermentation. Usually however, the non-flocculable cells soon pass into a flocculable condition (represented by shaded circles). While they remain in a non-flocculent environment however, the culture remains dispersed and flocs only form when the environment surrounding the flocculable cells becomes itself 'flocculent' as by progressive fermentation.

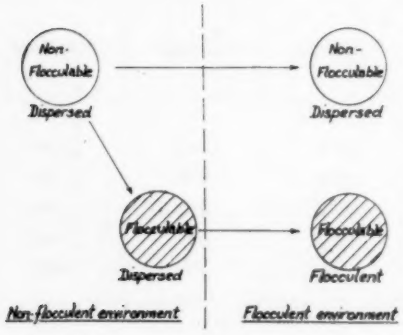


FIGURE 32. *Diagrammatic representation of factors controlling the flocculation of yeast*

CONSIDERATIONS REGARDING THE USE OF MIXED STRAINS OF YEAST

Many of the foregoing remarks bear closely on the behaviour of mixed strains of yeast, for in such circumstances the observed brewing behaviour is the resultant of the contributions of the individual strains. The individual contribution presumably depends on the proportion of the strain concerned. When a single strain is employed its properties appear to be constant, apart from rare variations due to mutation or segregation of characters arising from sporulation and rediploidization. On the other hand, a change in the proportions of such invariant strains in a mixed yeast can appear in practice to lead to a general change in character; and there is little doubt that such a change in proportions is not uncommon both from fermentation to fermentation and within individual fermentations. Where it does not take place, it is presumably because the empirical experience of the brewer has taught him when to skin off the yeast so as to ensure a reproducible fermentation. Where it does occur it may lead to imbalance or even ultimately to the suppression of one or more of the original strains. An illustration of such behaviour is afforded by Figure 33 which summarizes the composition of a fresh pitching yeast actually employed in a brewery and the composition of the yeast derived from it after several fermentation cycles.

Yeast	*Number out of thirty representative colonies having flocculence of:*			
		+++	++	+
Fresh pitching yeast ...	Colony type	0 —	15 15S	15 14R, 1S
After four fermentation cycles	Colony type	1 R	5 3R, 2S	24 21R, 3S

(R = rough, S = smooth)

FIGURE 33. *Progressive decrease of flocculence in a brewery yeast*

It will be seen first that the number of rough and of smooth colonies observed among thirty sample colonies is very different in the two cases. There seems usually to be a correspondence between roughness of colony character and non-flocculence and indeed, in the present case, when these thirty sample cultures were examined for flocculent behaviour it became clear that the flocculent cells were less numerous after use in the brewery. It was, in fact, a difficulty of drifting attenuation which had led to the examination of this yeast, with the significant result that it was confirmed that the yeast as such had not changed, but that the more flocculent strain or strains were being outgrown by the less flocculent ones. The opposite behaviour has also been observed of a mixed yeast appearing to become more flocculent owing to the flocculent strains being favoured on continued use.

CONCLUSION

The foregoing account may be said to bring one to the present frontiers of science as they impinge upon brewing. To reach these frontiers during only three lectures it has been necessary to traverse much important territory with often scarcely a glance at its notable landmarks. This has been inevitable in view of the many sciences with which brewing has contact and of the very wide field over which brewing research necessarily ranges. The results of this far-reaching work are, it is submitted, ample justification for carrying it out. As such efforts gain momentum both in this country and abroad, it is not too much to hope that not only brewers but all industry, and in addition science in general, will benefit to an increasing extent from the accumulated insight into the fascinating phenomena associated with brewing.

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GENERAL NOTES

POST-WAR CHURCH BUILDINGS EXHIBITION

A most interesting Exhibition devoted to developments in church architecture since the war was opened by the President of the Royal Academy, in the presence of the Archbishop of Canterbury at the Building Centre on 6th February. As the building of churches is a matter for each diocese, the Central Council for the Care of Churches organized this Exhibition in the hope that it would enable both architects and diocesan authorities to see what is being done in this field in different parts of the country.

The Exhibition, which closes on 17th February, is made up of photographs, plans and models. One interesting development which was made apparent in the Exhibition is the trend for the combination of the church and the church hall, either as one room which can be adapted for either use, or by the employment of sliding doors between the two.

EXHIBITION OF PICTURES FOR SCHOOLS

The ninth annual Exhibition of Pictures for Schools, organized by the Society for Education through Art, is at present on view at the Whitechapel Art Gallery, where it will remain until 4th March. The Exhibition contains works of art, in several media, which have been specially chosen as suitable for schools. The Exhibition is open on weekdays, excluding Mondays, from 11 a.m. to 6 p.m., and from 2 to 6 p.m. on Sundays. Admission is free.

TEXTILE DESIGN COMPETITION

Wool Review is again this year organizing a Competition for design of wool textiles. Prizes totalling 85 guineas are to be awarded in each of two groups: men's wear fabrics, and women's wear fabrics. A prize of 25 guineas is also being offered for the best tie fabric design, and one of twenty guineas for scarves. A prize of ten guineas will be donated for the best design in pure worsted fabric. Full details and entry forms may be obtained from *Wool Review*, 222 Strand, W.C.2. The closing date for applications is 30th April, 1956.

OBITUARY

MR. S. R. HOBDAY

We record with regret the death, in Sussex on 4th February, of Mr. S. R. Hobday.

Stephen Reginald Hobday, O.B.E., F.S.A., was born in 1875, and educated at London University, being called to the bar in 1901. He played a prominent part in the waterways industry throughout his life, being, before his retirement in 1948, Clerk of the Board and General Manager of the Lee Conservancy Board, and Clerk of the Lee Conservancy Catchment Board. During the last war he was a Member of the Central Canal Committee, and he was at one time Chairman of the National Joint Council for the Inland Waterways Industry. He took a keen interest in the prevention of pollution of rivers, and the recommendations of a sub-committee, of which he was chairman, led to the passing of the Rivers (Prevention of Pollution) Act of 1951. In 1935 he took the chair at a meeting at which a paper on 'Canals' was read to the Society by Mr. W. H. Curtis.

Mr. Hobday was elected a Fellow of the Society in 1936.

NOTES ON BOOKS

THE TEACH YOURSELF HISTORY OF PAINTING. *Based on the original work of H. Schmidt Degener, and edited for English readers by William Gaunt. Ten volumes. London, English Universities Press, 1954-5. 10s 6d each volume.*

Our generation has seen a greatly increased popular interest in painting and in the great masters and masterpieces of the past. It is not surprising, therefore, that that interest should show itself in a spate of books on the history of painting and, in particular, in a large number of monographs, or series of monographs, on famous painters, so that a knowledge which was formerly the possession of a few specially interested people has now been made available, in a superficial form at least, to a much wider public. One danger at least presents itself in this; that in the popular mind the great names become isolated from the tradition of which they were a part. The first value of a series such as this, therefore, which sets out to convey the whole range of European painting, is that it does in fact give, within the limits of almost exactly fifty pages each of ten volumes, a concise, if necessarily short, survey of its immense subject, and enables the general reader to put what is perhaps a fragmentary knowledge into a wider perspective. It does this in a text which is very readable, though not entirely free from the *clichés* of art history, and is critical whilst remaining objective. The reader is helped by a liberal choice of reproductions, which are well chosen to illustrate the argument and admirably associated with the text in format.

I imagine that this series of books will appeal most to the general reader who starts with an interest, which may be only partially informed, about the painters of the past, and there is little either in the factual material, or in the critical assessment of individual painters, which is new. The writer and editor have, on the whole, recorded the accepted verdict of contemporary criticism, but with careful accuracy and a knowledge of recent opinion on some controversial issues. There are, of course, points on which some readers may wish to differ—the rather curt dismissal of Sir Peter Lely as inferior to William Dobson, for instance—and perhaps a few passages which are misleading, as the reference to Tiepolo as 'the last great *fresco* painter among the Venetians', a school most of the greatest achievements of which were not in fresco. But the refusal of the authors to be deflected from a clear factual account to enter into purely personal opinions, which is a necessary virtue of a book in which references to individual painters must perforce be brief, gives to this history an added authority.

The ten volumes follow a conventional arrangement of the history of European painting. There are eight volumes devoted to the national schools, two for the Italians and one each for the Flemish, the Spanish, the German, the Dutch, the French and the British Schools, the ninth and tenth volumes treating the nineteenth and twentieth centuries respectively in their entirety. There is much to recommend this easy arrangement of an extremely complicated story. But, reading again through the story of European art, one is struck by the amazing complexity of it, and how closely interwoven are the manners and styles of painting in the various schools, which this series treats nationally. Until the end of the nineteenth century, when the flood gates were opened, European painting was fairly well insulated against outside influences, but within its own not narrow borders it is hard to put a limit to the degree to which ideas and influences were shared between painters of various nationalities. So many of the great masters were, in terms of art, European before they were Spanish or French or German. Travel between countries was common practice throughout the story, even in the sixteenth and seventeenth centuries. Consider, for instance, the Spanish School—if the term 'school' is not in itself misleading. No national influence was as strong with any of the great Spanish masters as outside European influences. Ribera, in the matters which concern a painter, was more a figure of the seventeenth century in Italy than a Spaniard. What Spanish link is there between the art of

El Greco and Velazquez? Can the gap in the history of Spanish painting between Velazquez and Goya be accounted for in the work of Spanish painters? Poussin and Claude must perforce be included in a volume on the French School, but they more truly belonged to, and influenced, a Roman School. It is more reasonable to speak of a 'school' of seventeenth century Dutch painters, or of the Pre-Raphaelites as a school. The authors seem to be aware of this handicap in the system of arrangement and have added at the end of each volume a 'Time Chart', relating the narrative of each school to contemporary events and painters, but one is left wondering whether the history of European painting can be so neatly fitted into seven schools, each, except one, of exactly fifty pages.

The volumes are attractively produced with good monochrome, and not quite so good colour, reproductions. It would have helped the reader, since appreciation of scale is an essential part of the understanding of a picture, had the dimensions of the originals been given. He is also left wishing for an index. But these are volumes warmly to be recommended to the ever-growing interest in the history of European painting.

DAVID BELL

SHORT NOTES ON OTHER BOOKS

THE RUDIMENTS OF LITHOGRAPHY. *By T. E. Griffits. Faber*, 1956. 12*s* 6*d*

Lithography, discovered in 1796, has been practised in London since 1823, but its recent return to favour has owed much to the work of the author of this book. His explanation of the craft is illustrated with many diagrams and half-tone plates.

CEZANNE (SECOND VOLUME). *Introduction and notes by Alan Clutton-Brock. Faber*, 1955. 12*s* 6*d*

Pictures now in public galleries and private collections in several countries are reproduced in this addition to the Faber gallery series. There is an introduction, and a note on each picture.

PAUL NASH: THE PORTRAIT OF AN ARTIST. *By Anthony Bertram. Faber*, 1955. 42*s*

The phases of Nash's work as an artist form the basis for the divisions into which this book falls. Each of the chapters begins with the chronicle of his life during the relevant years and then has sections on his work. There are 32 illustrations, a copious bibliography, and an index of pictures, as well as a general index.

FROM THE JOURNAL OF 1856

VOLUME IV. 15*th February*, 1856

From a paper On the Manufacture of Crown and Sheet Glass

I turn, in conclusion, . . . to . . ., the great ends which, from small beginnings, this manufacture has accomplished. The glazier of Pompeii little dreamt that the time would come when the substance that he handled, then so rare, would minister through the world to the health and pleasure of mankind; that beneath it trees would flourish, transported from a foreign soil; and that, cheaper than wood itself, it would combine with iron to form the largest building on the earth. But from Pompeii to the Crystal Palace there has been a progress of nearly twenty centuries, and a still greater progress in action than in time. That progress has been marked by no sudden revolutions, such as have distinguished the development of other efforts of mankind. The problem of blowing glass into a sheet admits of limited solutions, a cylinder or a sphere. Through one or other of these forms the first blown sheet cannot but have passed, and through one or other all pass now. But the progress has been none the less for lack of revolutionising discoveries. Minute and apparently unimportant changes, perceptible only to those near at hand, have in silence done their work.

Still, the development both of its processes and its applications is incomplete. Sir Joseph Paxton's glass arcade, stretching over the streets of London, and Dr. Sleigh's city of glass, in which invalids will inhale the gentle air of Madeira,—these are faint foreshadowings of the destiny of this wonderful substance.

And, perhaps, the day may come when some future chronicler of glass shall smile at the barbarians of the nineteenth century, who could not make window glass without defects, and shall exhibit those defects as the curiosities of a byegone age.

Some Activities of Other Societies and Organizations

MEETINGS

MON. 20 FEB. Electrical Engineers, Institution of, Savoy Place, W.C.2. 5.30 p.m. C. F. Brocklesby: *Ultrasonics in Industry.*

Geographical Society, Royal, South Kensington, S.W.7. 8.30 p.m. Dr. D. J. Crisp: *The Changing Climate of Northern Europe.*

Imperial Institute, South Kensington, S.W.7. 5.45 p.m. A. D. Azhar: *My Country: Pakistan.*

TUES. 21 FEB. Industrial Transport Association, at the Royal Society of Arts, W.C.2. 6.30 p.m. C. Courtney Cramp: *Some Transport Queries and the Answers.*

Manchester Geographical Society, 16 St. Mary's Parsonage, Manchester, 3. 6.30 p.m. Miss Jean Carter: *Uplands of the Cameroons.*

Mechanical Engineers, Institution of, 1 Birdcage Walk, S.W.1. 6.45 p.m. *The Influence of Appearance on Vehicle Design* (Discussion).

Photographic Society, Royal, 16 Princes Gate, S.W.7. 7 p.m. Frank W. Bryson: *Sixty Years of Marine Photography.*

Textile Institute, at 10 Blackfriars Street, Manchester, 3. 7 p.m. E. J. Davies: *The Dry Cleaner and You.* At the Midland Hotel, Bradford. 7.15 p.m. K. G. Ponting: *British Dominance in Wool.*

WED. 22 FEB. Central Asian Society, Royal, at the Royal Society, Burlington House, Piccadilly, W.1. 1.30 p.m. Major-General W. E. V. Abraham: *Burma.*

Electrical Engineers, Institution of, Savoy Place, W.C.2. 5.30 p.m. (1) B. C. Robinson: *The Penetration of Surge Voltages through a Transformer coupled to an Alternator.* (2) *The Propagation of Surge Voltages through Turbo-Alternators with Concentric Conductor Type Windings.*

Victoria & Albert Museum, South Kensington, S.W.7. 6.15 p.m. Margaret Medley: *Chinese Bird and Flower Painting.*

THURS. 23 FEB. Incorporated Plant Engineers, at the College of Preceptors, Bloomsbury Square, W.C.1. 7 p.m. D. C. Bean: *Pile driving plant— its use and maintenance.*

Physical Society, at the National Hospital, Queen Square, W.C.1. 5.30 p.m. D. E. Broadbent: *Physiological and Psychological Effects of Noise.*

Refrigeration, Institute of, at the Institution of Mechanical Engineers, 1 Birdcage Walk, S.W.1. 5.30 p.m. H. Burley and E. J. Perry: *Modern Trends in Dairy Refrigeration.*

SAT. 25 FEB. Horniman Museum, London Road, Forest Hill, S.E.23. 3.30 p.m. Dr. Marian W. Smith: *The Swaixwe: a social dance of the American Indians of the Pacific Coast.*

MON. 27 FEB. Imperial Institute, South Kensington, S.W.7. 5.45 p.m. Michael Newman: *My Country: The Federation of Rhodesia and Nyasaland.*

TUES. 28 FEB. Electrical Engineers, Institution of, Savoy Place, W.C.2. 5.30 p.m. M. W. Makowski and K. Mochlinski: *An Evaluation of Two Rapid Methods of Assessing the Thermal Resistivity of Soil.*

Illuminating Engineering Society, at the Lighting Service Bureau, 2 Savoy Hill, W.C.2. 6 p.m. K. Sutton-Jones: *Lighting Aids for Marine Navigation.*

Manchester Geographical Society, 16 St. Mary's Parsonage, Manchester, 3. 6.30 p.m. Sam Hanna: *Switzerland, Mediæval and Modern.*

WED. 29 FEB. Analytical Chemistry, Society for, at the Royal Society, Burlington House, Piccadilly, W.1. 5 p.m. Sir William Slater: *The Evolution of Agricultural Research.*

British Kinematograph Society, at the Holborn Town Hall, High Holborn, W.C.1. 7.15 p.m. P. H. Dorté: *The Building of the Independent Television News Service.*

Locomotive Engineers, Institution of, at the Institution of Mechanical Engineers, 1 Birdcage Walk, S.W.1. 5.30 p.m. Sir John D. Cockcroft: *Atomic Energy and Propulsion.*

Petroleum, Institute of, at 26 Portland Place, W.1. 6 p.m. W. Kohring: *Petroleum Refining in the United Kingdom.*

Victoria & Albert Museum, South Kensington, S.W.7. 6.15 p.m. Francis H. Taylor: *Modern American Art.*

THURS. 1 MAR. Anthropological Institute, Royal, 21 Bedford Square, W.C.1. 5.30 p.m. D. J. Stenning: *Migration: Migratory Drift and Transhumance Patterns of Fulani Nomadism.*

Textile Institute, at Cardiff University. 7.15 p.m. C. S. Whewell: *Modern Textile Finishing.*

FRI. 2 MAR. Mechanical Engineers, Institution of, 1 Birdcage Walk, S.W.1. 6.45 p.m. *British Standard Code for Acceptance Tests on Steam Generating Units* (Discussion).

SAT. 3 MAR. British Interplanetary Society, at Caxton Hall, Caxton Street, S.W.1. 6 p.m. Prof. A. D. Baxter: *Combustion Chambers for Rocket Engines.*

Horniman Museum, London Road, Forest Hill, S.E.23. 3.30 p.m. D. Chilton: *The Weather Glass.*

OTHER ACTIVITIES

NOW UNTIL 29 FEB. Victoria & Albert Museum, South Kensington, S.W.7. *Exhibition of Anglo-Jewish Art and History* (1656-1956).

NOW UNTIL 25 MAR. Imperial Institute, South Kensington, S.W.7. Exhibition: *Nigeria Welcomes the Queen.*

MON. 20 FEB. UNTIL SUN. 26 FEB. Imperial Institute, South Kensington, S.W.7. 12.30 p.m., 1.15 p.m. and 3 p.m. Weekdays. 3 p.m. and 4 p.m. Saturdays. 3 p.m., 4 p.m. and 5 p.m. Sundays. Films: *This is Nigeria; Northland Tour—New Zealand.*

WED. 22 FEB. The Building Centre, 26 Store Street, W.C.1. 12.45 p.m. Film Show: *Power Tools in the Building Industry.*

MON. 27 FEB. UNTIL SUN. 4 MAR. Imperial Institute, South Kensington, S.W.7. 12.30 p.m., 1.15 p.m. and 3 p.m. Weekdays. 3 p.m. and 4 p.m. Saturdays. 3 p.m., 4 p.m. and 5 p.m. Sundays. Films: *Australian Diary; Rhodesia Spotlight; The Rival World.*

WED. 29 FEB. The Building Centre, 26 Store Street, W.C.1. 12.45 p.m. Film Show: *Colt Canadian Cedarwood Shingles—Methods of Fixing.*